Hines
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- key terms

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     FILE 'REGISTRY' ENTERED AT 14:08:15 ON 16 SEP 1999
                E PHOSPHATIDYLINOSITOL 3 KINASE/CN 5
L1              1 S E4

     FILE 'CAPLUS' ENTERED AT 14:08:53 ON 16 SEP 1999

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L1              1 SEA FILE=REGISTRY ABB=ON  PLU=ON  "PHOSPHATIDYLINOSITOL
                  3'-KINASE"/CN
L2          13140 SEA FILE=CAPLUS ABB=ON  PLU=ON  L1 OR (PI3 OR (PHOSPHATID
                  YLINOSITOL OR PHOSPHATIDYL INOSITOL)(W)3)(W)KINASE OR
                  PHOSPHOTASE INOSITIDE OR PHOSPHOINOSIT? OR PHOSPHO(W)INOS
                  IT?
L3           7250 SEA FILE=CAPLUS ABB=ON  PLU=ON  L2 AND (POLYPEPTIDE OR
                  POLYPROTEIN OR PEPTIDE OR PROTEIN)
L4            102 SEA FILE=CAPLUS ABB=ON  PLU=ON  L3 AND (PCR OR POLYMERASE
                  (1W)REACT?)
L5             48 SEA FILE=CAPLUS ABB=ON  PLU=ON  L4 AND (DNA OR DEOXYRIBON
                  UCLEIC OR RNA OR RIBONUCLEIC OR NUCLEIC)
L8             32 SEA FILE=CAPLUS ABB=ON  PLU=ON  L5 AND GENE

=> d 1-32 .bevstr

L8   ANSWER 1 OF 32  CAPLUS  COPYRIGHT 1999 ACS
ACCESSION NUMBER:         1999:271498  CAPLUS
DOCUMENT NUMBER:          130:292101
TITLE:                    Novel isoforms of the human HER4/ErbB4 epidermal
                          growth factor receptors
INVENTOR(S):              Klagsbrun, Michael; Elenius, Klaus; Corfas,
                          Gabriel
PATENT ASSIGNEE(S):       Children's Medical Center Corporation, USA
SOURCE:                   PCT Int. Appl., 93 pp.
                          CODEN: PIXXD2
DOCUMENT TYPE:            Patent
LANGUAGE:                 English
FAMILY ACC. NUM. COUNT:   1
PATENT INFORMATION:

     PATENT NO.        KIND  DATE           APPLICATION NO.  DATE
     ---------------   ----  --------       ---------------  --------
     WO 9919488         A1   19990422       WO 1998-US21828  19981015
         W:  AU, CA, JP
         RW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC,
             NL, PT, SE
     AU 9898053         A1   19990503       AU 1998-98053    19981015
PRIORITY APPLN. INFO.:                     US 1997-62373    19971015
                                           WO 1998-US21828  19981015
AB   Novel isoforms of the human epidermal growth factor receptor 4 that
                          Searcher   :    Shears   308-4994
```

arise from differential splicing of the primary transcript are described. Two of the isoforms have 13 or 23 amino acids inserted in the juxtamembrane domain (the extracellular domain just N-terminal to the transmembrane domain). Two isoforms affecting the cytoplasmic domain are also reported, one is a deletion variant lacking 15 amino acids of the cytoplasmic tail, including a binding site for the p85 subunit of **phosphatidylinositol 3 -kinase** and does not stimulate the kinase. These isoforms have specific tissue distributions. The cDNAs were identified in mouse by sequencing of **PCR** amplification products from various tissues using primers derived from a previously published sequence. The **proteins** retained binding of neuregulins and other growth factors and were inactivated by phorbol esters.

IT **115926-52-8, Phosphatidylinositol 3-kinase**
RL: BPR (Biological process); BIOL (Biological study); PROC (Process)
(HER4/ErbB4 isoforms interaction with; novel isoforms of human HER4/ErbB4 epidermal growth factor receptors)

L8 ANSWER 2 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1999:187459 CAPLUS
DOCUMENT NUMBER: 131:14398
TITLE: Pituitary adenylate cyclase-activating **polypeptide** modulates gastric enterochromaffin-like cell proliferation in rats
AUTHOR(S): Lauffer, Jorg M.; Modlin, Irvin M.; Hinoue, Toshinori; Kidd, Mark; Zhang, Tong; Schmid, Stefan W.; Tang, Laura H.
CORPORATE SOURCE: Gastric Pathobiology Research Group, Department of Surgery, Yale University School of Medicine and West Haven Veterans Administration Medical Center, New Haven, CT, USA
SOURCE: Gastroenterology (1999), 116(3), 623-635
CODEN: GASTAB; ISSN: 0016-5085
PUBLISHER: W. B. Saunders Co.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Gastric carcinoids (types I and II) involve the transformation of naive enterochromaffin-like (ECL) cells to the neoplastic state and are assocd. primarily with hypergastrinemia. In this study, the authors evaluated the effects of two related neuropeptides, pituitary adenylate cyclase-activating **polypeptide** (PACAP) and vasoactive intestinal **polypeptide** (VIP), on ECL cell proliferation and characterized the receptor subtype(s) and signal transduction pathways that mediate this effect. Purified rat ECL cells were analyzed in culture for **DNA** synthesis as measured by 24-h 5-bromo-2-deoxyuridine (BrdU) uptake. Reverse-transcription **polymerase** chain **reaction**

(RT-**PCR**) with **gene**-specific oligonucleotide primers was performed to characterize the PACAP/VIP receptor subtype(s). PACAP/VIP neuropeptide-stimulated BrdU uptake was significantly greater (3.4-3.8-fold greater than control) than that at the maximal dose of gastrin (2.2-fold greater than control). PACAP-stimulated ECL cell proliferation (EC50, .apprx.3 .times. 10-14 M) was .apprx.100-fold more potent than VIP (EC50, .apprx.3 .times. 10-12 M). The stimulated BrdU uptake by both PACAP and VIP was competitively inhibited by PACAP-receptor antagonist (IC50, 10-9 M, 3 .times. 10-9 M, resp.) and VIP-receptor antagonist (IC50, 3 .times. 10-7 M, 5 .times. 10-7 M, resp.). RT-**PCR** identified the presence of the PACAP-specific but not PACAP/VIP receptor subtypes. The PACAP-stimulated BrdU uptake was inhibited (70%-80%) by inhibitors of cAMP, **phosphatidylinositol 3 kinase**, and **protein** tyrosine kinase as well as mitogen-activated **protein** kinase. PACAP/VIP-related **peptides** are more potent modulators of ECL cell proliferation than gastrin, and their effect is mediated by a PACAP-specific receptor whose activation is transduced by multiple intracellular messenger systems.

IT **115926-52-8, Phosphatidylinositol-3 kinase**
RL: BPR (Biological process); BIOL (Biological study); PROC (Process)
(PACAP and VIP signaling mechanism in modulation of gastric enterochromaffin-like cell proliferation)

L8 ANSWER 3 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1999:96378 CAPLUS
DOCUMENT NUMBER: 130:164014
TITLE: **Genes** controlling phytate metabolism enzymes in plants and their uses to control phytic acid or non-phytic phosphorus levels
INVENTOR(S): Martino-Catt, Susan J.; Wang, Hongyu; Beach, Larry R.; Bowen, Benjamin A.; Wang, Xun
PATENT ASSIGNEE(S): Pioneer Hi-Bred International, Inc., USA
SOURCE: PCT Int. Appl., 86 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9905298	A1	19990204	WO 1998-US14657	19980717

W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, GM, HR, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK,

MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL,
TJ, TM, TR, TT, UA, UG, US, US, US, US, US, UZ, VN, YU, ZW,
AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, CY, DE, DK,
ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF,
CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

AU 9884875 A1 19990216 AU 1998-84875 19980717
PRIORITY APPLN. INFO.: US 1997-53371 19970722
US 1997-53944 19970728
US 1997-55526 19970808
US 1997-55446 19970811
US 1998-85852 19980518
WO 1998-US14657 19980717

AB This invention relates to newly identified polynucleotides and **polypeptides**, variants and derivs. of same; methods for making the polynucleotides, **polypeptides**, variants, derivs. and antagonists. In particular the invention relates to polynucleotides and **polypeptides** of the phytate metabolic pathway. **Nucleic** acid and deduced amino acid sequences are provided for **phosphatidylinositol 3-kinase**, myo-inositol 1,3,4-triphosphate 5/6 kinase, myo-inositol monophosphatase-3, and myo-inositol 1-phosphate synthase from Zea mays. Primers to amplify these **genes** from a Zea mays **nucleic** acid library are also provided. These **genes** can be used to provide transgenic plants with decreased levels of phytic acid or increased non-phytic phosphorus, as well as improving animal performance with feeds contg. transformed plant materials.

IT **115926-52-8, Phosphatidylinositol 3-kinase**
RL: BOC (Biological occurrence); BUU (Biological use, unclassified); PRP (Properties); BIOL (Biological study); OCCU (Occurrence); USES (Uses)
(**genes** controlling phytate metab. enzymes in plants and their uses to control phytic acid or non-phytic phosphorus levels)

L8 ANSWER 4 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1999:95663 CAPLUS
DOCUMENT NUMBER: 130:250555
TITLE: Mutational analysis of the coding regions of the **genes** encoding **protein** kinase B-.alpha. and -.beta., **phosphoinositide** -dependent **protein** kinase-1, phosphatase targeting to glycogen, **protein** phosphatase inhibitor-1, and glycogenin: lessons from a search for genetic variability of the insulin-stimulated glycogen synthesis pathway of skeletal muscle in NIDDM

patients

AUTHOR(S): Hansen, Lars; Fjordvang, Helle; Rasmussen, Soren K.; Vestergaard, Henrik; Echwald, Soren M.; Hansen, Torben; Alessi, Dario; Shenolikar, Shirish; Saltiel, Alan R.; Barbetti, Fabrizio; Pedersen, Oluf

CORPORATE SOURCE: Steno Diabetes Center and Hagedorn Research Institute, Gentofte, DK-2820, Den.

SOURCE: Diabetes (1999), 48(2), 403-407
CODEN: DIAEAZ; ISSN: 0012-1797

PUBLISHER: American Diabetes Association

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The finding of a reduced insulin-stimulated glucose uptake and glycogen synthesis in the skeletal muscle of glucose-tolerant first-degree relatives of patients with NIDDM, as well as in cultured fibroblasts and skeletal muscle cells isolated from NIDDM patients, has been interpreted as evidence for a genetic involvement in the disease. The mode of inheritance of the common forms of NIDDM is as yet unclear, but the prevailing hypothesis supports a polygenic model. In the present study, the authors tested the hypothesis that the putative inheritable defects of insulin-stimulated muscle glycogen synthesis might be caused by genetic variability in the **genes** encoding **proteins** shown by biochem. evidence to be involved in insulin-stimulated glycogen synthesis in skeletal muscle. In 70 insulin-resistant Danish NIDDM patients, mutational anal. by reverse transcription-**polymerase** chain **reaction**-single strand conformation polymorphism-heteroduplex anal. was performed on genomic **DNA** or skeletal muscle-derived cDNAs encoding glycogenin, **protein** phosphatase inhibitor-1, phosphatase targeting to glycogen, **protein** kinase B-.alpha. and -.beta., and the **phosphoinositide**-dependent **protein** kinase-1. Although a no. of silent variants were identified in some of the examd. **genes**, the authors found no evidence for the hypothesis that the defective insulin-stimulated glycogen synthesis in skeletal muscle in NIDDM is caused by structural changes in the **genes** encoding the known components of the insulin-sensitive glycogen synthesis pathway of skeletal muscle.

L8 ANSWER 5 OF 32 CAPLUS COPYRIGHT 1999 ACS

ACCESSION NUMBER: 1999:87395 CAPLUS

DOCUMENT NUMBER: 130:280242

TITLE: Characterization of ATM **gene** mutations in 66 ataxia telangiectasia families

AUTHOR(S): Sandoval, Natalia; Platzer, Matthias; Rosenthal, Andre; Dork, Thilo; Bendix, Regina; Skawran, Britta; Stuhrmann, Manfred; Wegne, Rolf-Dieter;

Sperling, Karl; Banin, Sharon; Shiloh, Yosef; Baumer, Alessandra; Bernthaler, Ulrike; Sennefelde, Helga; Brohm, Monika; Weber, Bernhard H. F.; Schindler, Detlev
CORPORATE SOURCE: Department of Genome Analysis, Institute of Molecular Biotechnology, Jena, Germany
SOURCE: Hum. Mol. Genet. (1999), 8(1), 69-79
CODEN: HMGEE5; ISSN: 0964-6906
PUBLISHER: Oxford University Press
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Ataxia telangiectasia (AT) is an autosomal recessive disease characterized by neurol. and immunol. symptoms, radiosensitivity and cancer predisposition. The **gene** mutated in AT, designated the ATM **gene**, encodes a large **protein** kinase with a PI-3 kinase-related domain. In this study, we investigated the mutational spectrum of the ATM **gene** in a cohort of AT patients living in Germany. We amplified and sequenced all 66 exons and the flanking untranslated regions from genomic **DNA** of 66 unrelated AT patients. We identified 46 different ATM mutations and 26 sequence polymorphisms and variants scattered throughout the **gene**. A total of 34 mutations have not been described in other populations. Seven mutations occurred in more than one family, but none of these accounted for more than five alleles in our patient group. The majority of the mutations were truncating, confirming that the absence of full-length ATM **protein** is the most common mol. basis of AT. Transcript analyses demonstrated single exon skipping as the consequence of most splice site substitutions, but a more complex pattern was obsd. for two mutations. Immunoblot studies of cell lines carrying ATM missense substitutions or in-frame deletions detected residual ATM **protein** in four cases. One of these mutations, a valine deletion proximal to the kinase domain, resulted in ATM **protein** levels >20% of normal in an AT lymphoblastoid cell line. In summary, our results survey and characterize a plethora of variations in the ATM **gene** identified by exon scanning sequencing and indicate a high diversity of mutations giving rise to AT in a non-isolated population.
IT **115926-52-8, Phosphatidylinositol 3-kinase**
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(putative, **gene** ATM **protein** kinase; characterization of ATM **gene** mutations in 66 ataxia telangiectasia families)

L8 ANSWER 6 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1998:763241 CAPLUS
DOCUMENT NUMBER: 130:136967
TITLE: .gamma.-Aminobutyric acid up-regulates the

expression of a novel secretogranin-II messenger **ribonucleic** acid in the goldfish pituitary

AUTHOR(S): Blazquez, Mercedes; Bosma, Peter T.; Chang, John P.; Docherty, Kevin; Trudeau, Vance L.

CORPORATE SOURCE: Department of Zoology, University of Aberdeen, Aberdeen, AB24 2TZ, UK

SOURCE: Endocrinology (1998), 139(12), 4870-4880
CODEN: ENDOAO; ISSN: 0013-7227

PUBLISHER: Endocrine Society

DOCUMENT TYPE: Journal

LANGUAGE: English

AB An **RNA**-arbitrarily primed **PCR** differential display strategy was used to identify candidate **genes** in the pituitary that are up-regulated by endogenously activated GABA systems that may also be involved in the control of reprodn. Goldfish were injected with the GABA metab. inhibitor .gamma.-vinyl-GABA (GVG), known for its high efficiency to specifically increase endogenous brain and pituitary GABA levels in this species, resulting in higher levels of circulating gonadotropin-II (GTH-II). Several transcripts related to hormone secretion, signal transduction pathways, and mRNA editing were shown to be up-regulated after GVG injection. Among these transcripts we characterized an mRNA coding for the secretory vesicle **protein** secretogranin-II (SgII), a member of the chromogranin family, which is the precursor of a novel 34 amino acid neuropeptide, goldfish secretoneurin (SN). A semiquant. **PCR** developed to measure pituitary SgII mRNA levels showed a 5-fold increase in GVG treated fish vs. control fish. Moreover, GVG treatment specifically increased SgII mRNA levels in gonadotrophs, concomitant with a decrease in GTH-II cell content. In addn., i.p. injection of synthetic goldfish SN increased GTH-II release in goldfish pretreated with the dopamine antagonist domperidone. Activation of GABAergic neurons has 2 effects, enhancing in vivo GTH-II release and up-regulating SgII mRNA specifically in goldfish gonadotrophs. Together with our SN bioactivity data, this suggests the existence in the pituitary of an autocrine or paracrine mechanism linked to the regulated secretory pathway in the gonadotrophs.

IT **115926-52-8, Phosphatidylinositol 3-kinase**
RL: BPR (Biological process); MFM (Metabolic formation); PRP (Properties); BIOL (Biological study); FORM (Formation, nonpreparative); PROC (Process)
(GABA upregulates secretogranin-II mRNA expression in goldfish pituitary)

L8 ANSWER 7 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1998:761816 CAPLUS

DOCUMENT NUMBER: 130:29188
TITLE: Therapeutic and diagnostic tools for impaired glucose tolerance conditions based on the dauer **polypeptides** and **genes** of Caenorhabditis elegans
INVENTOR(S): Ruvkun, Gary; Kimura, Koutarou; Patterson, Garth; Ogg, Scott; Paradis, Suzanne; Tissenbaum, Heidi; Morris, Jason; Koweek, Allison; Pierce, Sarah
PATENT ASSIGNEE(S): The General Hospital Corporation, USA
SOURCE: PCT Int. Appl., 202 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9851351	A1	19981119	WO 1998-US10080	19980515
W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, GM, GW, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
AU 9874941	A1	19981208	AU 1998-74941	19980515
PRIORITY APPLN. INFO.:			US 1997-857076	19970515
			US 1997-888534	19970707
			WO 1998-US10080	19980515

AB Disclosed herein are novel **genes** and methods for the screening of therapeutics useful for treating impaired glucose tolerance conditions, as well as diagnostics and therapeutic compns. for identifying or treating such conditions. The Caenorhabditis elegans metabolic regulatory **genes** daf-2 and age-1 encode homologs of the mammalian insulin receptor/**phosphoinositide** 3-kinase signaling pathway **proteins**, resp. In addn., the DAF-16 forkhead **protein** represents the major transcriptional output of this insulin signaling pathway. Dysregulation of the DAF-16 transcription factor in the absence of insulin signaling leads to metabolic defects; inactivation of DAF-16 reverses the metabolic defects caused by lack of insulin signaling in C. elegans. Finally, the C. elegans daf-7, da-1, daf-4, daf-8, daf-14, and daf-3 **genes** encode neuroendocrine/target tissue transforming growth factor-.beta. type signal transduction mols. that genetically interact with the insulin signaling pathway.

Metabolic defects cause by lack of neuroendocrine TGF-.beta. signals can be reversed by inactivation of the DAF-3 transcription factor. The C. elegans daf **genes** are excellent candidate **genes** and **proteins** for human disease assocd. with glucose intolerance, e.g., diabetes, obesity, and atherosclerosis. The human homologs of these daf **genes** and **proteins** mediate insulin signaling in normal people and may be defective or mis-regulated in diabetics. Moreover, there are at least 2 classes of type II diabetics: those with defects in the TGF-.beta. signaling **genes**, and those with defects in insulin signaling **genes**. Exemplary sequences and functional characteristics are provided for the C. elegans daf homologs of the human **genes**: daf-2, daf-3 (3 differentially spliced isoforms), daf-16 (2 differentially spliced isoforms), age-1, and pdk-1 (two spliced isoforms).

IT **115926-52-8**, **Phosphoinositide** 3-kinase
RL: BPR (Biological process); BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study); PROC (Process)
(therapeutic and diagnostic tools for impaired glucose tolerance conditions based on the dauer **polypeptides** and **genes** of Caenorhabditis elegans)

L8 ANSWER 8 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1998:367861 CAPLUS
DOCUMENT NUMBER: 129:107306
TITLE: Inhibition by platelet-activating factor of Src- and hepatocyte growth factor-dependent invasiveness of intestinal and kidney epithelial cells. **Phosphatidylinositol 3'-kinase** is a critical mediator of tumor invasion
AUTHOR(S): Kotelevets, Larissa; Noe, Veerle; Bruyneel, Erik; Myssiakine, Evgueni; Chastre, Eric; Mareel, Marc; Gespach, Christian
CORPORATE SOURCE: INSERM U482 and IFR 65, Hopital Saint Antoine, Paris, 75571, Fr.
SOURCE: J. Biol. Chem. (1998), 273(23), 14138-14145
CODEN: JBCHA3; ISSN: 0021-9258
PUBLISHER: American Society for Biochemistry and Molecular Biology
DOCUMENT TYPE: Journal
LANGUAGE: English

AB This study was designed to characterize platelet-activating factor receptor (PAF-R) expression and function in normal and cancerous human colonic epithelial cells. PAF-R **gene** transcripts were analyzed by reverse transcription-**polymerase** chain **reaction** and Southern blot, using three sets of primers corresponding either to the coding region of the human PAF-R sequence (**polymerase** chain **reaction** product: 682

base pairs (bp)) or to the leukocyte- and tissue-type transcripts of 166 and 252 bp, resp. An elongated splice variant was identified in the 5'-untranslated region of the tissue-type PAF-R transcript (334 bp) in colonic epithelial crypts and tumors. In human colonic PCmsrc cells transformed by c-src oncogene, the hepatocyte growth factor (HGF)-dependent invasiveness of collagen gels was abolished by 0.1 .mu.M PAF and restored by the PAF-R antagonists WEB2086 and SR27417. PAF blocked HGF-induced tyrosine phosphorylation of p125 focal adhesion kinase. The **phosphatidylinositol 3 '-kinase** (PI3'-K) inhibitors wortmannin and LY294002 totally blocked the HGF-induced invasion. Similar effects were obsd. in ts-srcMDCK kidney epithelial cells transformed by a v-Src temp.-sensitive mutant: (i) PAF and wortmannin exerted additive inhibitory effects on Src-induced invasion and (ii) activated and dominant neg. forms of p110.alpha. PI3'-K, resp., amplified and abrogated the Src- and HGF-dependent invasiveness of parental and ts-src-MDCK cells. The authors also provided the first evidence for the contribution of rapamycin-insensitive, pertussis toxin-dependent G-**protein** pathways to the integration of the signals emerging from activated Met and PAF receptors. These results indicate that PI3'-K is a crit. transducer of invasiveness and strongly suggest that PAF exerts a neg. control on invasion by inhibiting this signaling pathway. A possible beneficial role of PAF analogs on tumor invasion is therefore proposed.

IT **115926-52-8, Phosphatidylinositol 3-kinase**
RL: ADV (Adverse effect, including toxicity); BAC (Biological activity or effector, except adverse); BPR (Biological process); BIOL (Biological study); PROC (Process)
(inhibition by platelet-activating factor of Src- and hepatocyte growth factor-dependent invasiveness of intestinal and kidney epithelial cells and **phosphatidylinositol 3'-kinase** as crit. mediator of tumor invasion)

L8 ANSWER 9 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1998:74268 CAPLUS
DOCUMENT NUMBER: 128:213944
TITLE: Genetic characterization of a phospholipase C **gene** from Candida albicans: presence of homologous sequences in Candida species other than Candida albicans
AUTHOR(S): Bennett, Desiree E.; McCreary, Christine E.; Coleman, David C.
CORPORATE SOURCE: School Dental Science, Department Oral Medicine Pathology, Trinity College, Department Microbiology, University Dublin, Moyne Institute Preventive Medicine, Dublin, Ire.
SOURCE: Microbiology (Reading, U. K.) (1998), 144(1), 55-72

CODEN: MROBEO; ISSN: 1350-0872
PUBLISHER: Society for General Microbiology
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Phospholipase C (PLC) enzymes are essential in regulating several important cellular functions in eukaryotes, including yeasts. In this study, **PCR** was used to identify a **gene** encoding PLC activity in Candida albicans, using oligonucleotide primers complementary to sequences encoding highly conserved amino acid regions within the X domains of previously characterized eukaryotic phospholipase C **genes**. The nucleotide sequence of the C. albicans **gene**, CAPLC1 (2997 bp), was detd. from a recombinant clone contg. C. albicans 132A genomic **DNA**; it encoded a **polypeptide** of 1099 amino acids with a predicted mol. mass of 124.cntdot.6 kDa. The deduced amino acid sequence of this **polypeptide** (CAPLC1) exhibited many of the features common to previously characterized PLCs, including specific X and Y catalytic domains. The CAPLC1 **protein** also exhibited several unique features, including a novel stretch of 18-19 amino acid residues within the X domain and an unusually long N-terminus which did not contain a recognizable EF-hand Ca2+-binding domain. An overall amino acid homol. of more than 27% with PLCs previously characterized from Saccharomyces cerevisiae and Schizosaccharomyces pombe suggested that the CAPLC1 **protein** is a .delta.-form of **phosphoinositide**-specific PLC (PI-PLC). PLC activity was detected in cell-free exts. of both yeast and hyphal forms of C. albicans 132A following 7 h and 24 h growth using the PLC-specific substrate p-nitrophenylphosphorylcholine (p-NPPC). In addn. CAPLC1 mRNA was detected by reverse transcriptase **PCR** in both yeast and hyphal forms of C. albicans 132A at the same time intervals. Expression of CAPLC1 activity was also detected in exts. of Escherichia coli DH5.alpha. harboring plasmids which contained portions of the CAPLC1 **gene** lacking sequences encoding part of the N-terminus. Southern hybridization and **PCR** analyses revealed that all C. albicans and Candida dubliniensis isolates examd. possessed sequences homologous to CAPLC1. Sequences related to CAPLC1 were detected in some but not all isolates of Candida tropicalis, Candida glabrata and Candida parapsilosis tested, but not in the isolates of Candida krusei, Candida kefyr, Candida guillermondii and Candida lusitaniae examd. This paper reports the first description of the cloning and sequencing of a PLC **gene** from a pathogenic yeast species.

L8 ANSWER 10 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1997:748078 CAPLUS
DOCUMENT NUMBER: 128:124323
TITLE: The Arabidopsis thaliana genome has multiple divergent forms of **phosphoinositol**

-specific phospholipase C
AUTHOR(S): Hartweck, Lynn M.; Llewellyn, Danny J.; Dennis, Elizabeth S.
CORPORATE SOURCE: GPO Box, Plant Industry, Industrial Research Organisation, Commonwealth Scientific, Canberra, ACT 2601, 1600, Australia
SOURCE: Gene (1997), 202(1/2), 151-156
CODEN: GENED6; ISSN: 0378-1119
PUBLISHER: Elsevier Science B.V.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Highly degenerate primers to conserved regions of the eukaryotic **phosphoinositol**-specific phospholipase C (PLC) were used to amplify fragments of plant PLCs from Arabidopsis thaliana genomic **DNA**. Eight completely different fragment sequences that showed high homol. to PLCs of both animals and plants were isolated. The variation between these putative PLCs was high and suggests that, like animals, plants have multiple isoforms of PLC. Using one of the **PCR** clones, a corresponding full-length Arabidopsis PLC **gene** (ATHATPLC1G) was isolated, and sequence anal. indicated that it was most like a delta-type PLC. This **gene** is 2.5 kb and contains seven introns, all but one of which has intron/exon border sequences that conform to the Arabidopsis consensus. The structural complexity of the **gene** is relatively simple compared to mammalian .beta.-type PLCs that can be 15 kb long with up to 30 introns. The plant **gene** is a single copy and was mapped to four Arabidopsis YACs, one located on chromosome 2. The promoter region contained two TATA-like elements at -43 and -185 and other putative regulatory elements that suggest that this PLC is hormonally regulated. This is the first plant PLC **gene** and the first delta type-PLC **gene** from a higher organism to be sequenced.

L8 ANSWER 11 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1996:727122 CAPLUS
DOCUMENT NUMBER: 126:27496
TITLE: Molecular cloning of the Toxoplasma gondii sag4 **gene** encoding an 18 kDa bradyzoite specific surface **protein**
AUTHOR(S): Oedberg-Ferragut, Carmen; Soete, Martine; Engels, Anne; Samyn, Bart; Loyens, Anne; Van Beeumen, Jozef; Camus, Daniel; Dubremetz, Jean-Francois
CORPORATE SOURCE: INSERM U42, 369, rue Jules Guesde, BP. 39, Villeneuve d'Ascq, 59651, Fr.
SOURCE: Mol. Biochem. Parasitol. (1996), 82(2), 237-244
CODEN: MBIPDP; ISSN: 0166-6851
PUBLISHER: Elsevier
DOCUMENT TYPE: Journal

LANGUAGE: English

AB An 18 kDa bradyzoite sp. surface **protein** of Toxoplasma gondii (T. gondii) has been purified by affinity chromatog. with a specific monoclonal antibody using parasites grown in vitro under conditions inducing the biosynthesis of bradyzoite specific **proteins**. N-terminal and internal amino acid sequences obtained by micro-sequencing enabled us to design degenerate oligonucleotides. A fragment of 187 bp was amplified by **polymerase** chain **reaction** (**PCR**). It was used as a probe to clone a 4 kb-BamHI fragment encompassing the **gene** encoding the 18 kDa **protein**. Nucleotide sequence anal. revealed a single open reading frame of 516 nucleotides encoding a 172 amino acid **protein**. The deduced amino acid sequence matched perfectly the **peptides** micro-sequenced from the native **protein**. The N-terminal hydrophobic region was found to possess the characteristics of a signal **peptide** of 27 amino acids. The hydrophobic C-terminal part could represent a signal for a glycan-**phosphoinositide** anchor. The full-length cDNA was also isolated and both the 5' and 3' untranslated regions were detd. Reverse transcriptase-**PCR** (RT-**PCR**) using p18-specific primers showed a stage-specific expression of this **gene**. Comparison of the **nucleic** acid sequence and the predicted amino acid sequence with databases did not reveal significant homol. with known **genes** or **proteins**. This **gene** is proposed to be named sag4, according to the existing T. gondii nomenclature.

L8 ANSWER 12 OF 32 CAPLUS COPYRIGHT 1999 ACS

ACCESSION NUMBER: 1996:655374 CAPLUS

DOCUMENT NUMBER: 126:1514

TITLE: Identification, molecular cloning, and distribution of a short variant of the 5-hydroxytryptamine2C receptor produced by alternative splicing

AUTHOR(S): Canton, Herve; Emeson, Ronald B.; Barker, Eric L.; Backstrom, Jon R.; Lu, Jonathan T.; Chang, Mike S.; Sanders-Bush, Elaine

CORPORATE SOURCE: Department Pharmacology, Vanderbilt University School Medicine, Nashville, TN, 37232, USA

SOURCE: Mol. Pharmacol. (1996), 50(4), 799-807
CODEN: MOPMA3; ISSN: 0026-895X

PUBLISHER: Williams & Wilkins

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The actions of the neurotransmitter 5-hydroxytryptamine (5-HT) (serotonin) are mediated by multiple receptor subtypes. One of the prominent serotonin receptors in the brain is the 5-HT2C receptor (5-HT2C-R). The authors report the occurrence of a second 5-HT2C-R

transcript, first identified using S1 nuclease protection of total **RNA** isolated from the choroid plexus. Analyses of the distribution of these two **RNAs** revealed that the short form is expressed in the same structures as the 5-HT2C-R mRNA, including choroid plexus, striatum, hippocampus, hypothalamus, olfactory tubercles, and spinal cord. Cloning and sequence analyses revealed a second cDNA with a 95-nt deletion in the region coding for the putative second intracellular loop and the fourth transmembrane domain of the 5-HT2C-R. This deletion leads to a frameshift in the coding sequence and the introduction of a premature stop codon. The predicted truncated **protein** (5-HT2C-tr) contains 172 amino acids, with 153 residues at the N-terminus, identical to the 5-HT2C-R, and 19 C-terminal amino acids that are unique. Although antibodies specific to the 5-HT2C-tr **protein** showed that the truncated form is expressed in a transfected fibroblast cell model system, there was no serotonergic ligand binding activity or **phosphoinositide** hydrolysis. Analyses of the 5-HT2C-R **gene** revealed that the two transcripts arise from a single **gene** by differential splicing using alternative donor sites and a common 3'-splice acceptor. **Polymerase** chain **reaction** amplification of mouse and human brain cDNAs demonstrated the occurrence of the same splicing patterns in these species. Although this study demonstrates tissue-specific expression of two 5-HT2C mRNA splice variants in rat, mouse, and human, the significance of the truncated form in these three species remains to be established.

L8 ANSWER 13 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1996:606459 CAPLUS
DOCUMENT NUMBER: 125:267057
TITLE: The Friedreich's ataxia **gene** encodes a novel phosphatidylinositol-4-phosphate 5-kinase
AUTHOR(S): Carvajal, Jaime J.; Pook, Mark A.; dos Santos, Maria; Doudney, Kit; Hillermann, Renate; Minogue, Shane; Williamson, Robert; Hsuan, J. Justin; Chamberlain, Susan
CORPORATE SOURCE: Hereditary Ataxia Research Group, Imperial College School Medicine St. Mary's, London, W2 1PG, UK
SOURCE: Nat. Genet. (1996), 14(2), 157-162
CODEN: NGENEC; ISSN: 1061-4036
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The STM7 **gene** on chromosome 9 was recently excluded as a candidate for Friedreich's ataxia following the identification of an expanded intronic GAA triplet repeat in the adjacent **gene**, X25, in patients with the disease. Using RT-**PCR**, Northern and sequence analyses, we now demonstrate that X25 comprises part of the STM7 **gene**, contributing to at least four splice

variants, and report the identification of new coding sequences. Functional anal. of the STM7 recombinant **protein** corresponding to the reported 2.7-kilobase transcript has demonstrated PtdInsP 5-kinase activity, supporting the idea that the disease is caused by a defect in the **phosphoinositide** pathway, possibly affecting vesicular trafficking or synaptic transmission.

L8 ANSWER 14 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1996:592576 CAPLUS
DOCUMENT NUMBER: 125:245488
TITLE: Biochemical and genetic defects in the **DNA**-dependent **protein** kinase in murine scid lymphocytes
AUTHOR(S): Danska, Jayne S.; Holland, Dianne P.; Mariathasan, Sanjeev; Williams, Kelly M.; Guidos, Cynthia J.
CORPORATE SOURCE: Division Surgical Research, University Toronto, Toronto, ON, Can.
SOURCE: Mol. Cell. Biol. (1996), 16(10), 5507-5517
CODEN: MCEBD4; ISSN: 0270-7306
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The scid **gene** product has been identified as the 460-kDa catalytic subunit of the **DNA**-dependent **protein** kinase (**DNA**-PKcs p460), a member of the **phosphatidylinositol 3-kinase** family. **DNA**-PK activity is undetectable in scid cells, but the mol. basis for this defect has not been identified. Here we report that expression of p460 in scid lymphocyte precursors is detectable but is reduced at least 10-fold relative to that in wild-type lymphocyte. In addn., we show that the scid mutation disturbs p460 nuclear assocn., presumably affecting its role in **DNA** repair pathways. To examine the mol. basis for our observations, we used a degenerate **PCR** strategy to clone the C-terminal p460 kinase domain from wild-type and scid thymocytes. Northern (**RNA**) anal. with these probes revealed normal steady-state p460 mRNA levels in scid cells, suggesting that the reduced abundance of p460 **protein** is due to a posttranscriptional defect. Sequence comparisons identified a single-base-pair alteration in the scid C-terminal p460 kinase domain, resulting in a premature stop codon. This mutation is predicted to truncate p460 by approx. 8 kDa, but it preserves the conserved motifs required for kinase activity in members of the **phosphoinositidyl** 3-kinase family. Despite a computed mol. wt. alteration of less than 2%, we were able to visualize this difference by Western blot (immunoblot) anal. of wild-type and scid p460. These data demonstrate that the scid **DNA**-PKcs mutation is not a null allele and suggest a mol. rationale for the well-described leakiness

of the scid phenotype.

L8 ANSWER 15 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1996:573005 CAPLUS
DOCUMENT NUMBER: 125:241492
TITLE: Cloning and expression of a human placenta inositol 1,3,4,5-tetrakisphosphate and phosphatidylinositol 3,4,5-trisphophate 5-phosphatase
AUTHOR(S): Drayer, A. Lyndsay; Pesesse, Xavier; De Smedt, Florence; Woscholski, Rudiger; Parker, Peter; Erneux, Christophe
CORPORATE SOURCE: Interdisciplinary Res. Inst., Universite Libre de Bruxelles, Brussels, 1070, Belg.
SOURCE: Biochem. Biophys. Res. Commun. (1996), 225(1), 243-249
CODEN: BBRCA9; ISSN: 0006-291X
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Distinct inositol and phosphatidylinositol polyphosphate 5-phosphatases have recently been cloned. Primers were designated coding for highly conserved amino acid regions that are shared between sequences of 5-phosphatases. The authors used degenerate primers to amplify **polymerase** chain **reaction** products from rat brain cDNA. A product with a novel sequence was identified and used to clone a 4.9 kb cDNA from human placenta cDNA libraries (hp51CN). COS-7 cells transfected with a C-terminal truncated form of this cDNA showed an increase in Ins(1,3,4,5)P4 and PtdIns(3,4,5)P3 hydrolyzing activity, but not in Ins(1,3,4,5)P3 5-phosphatase. Enzymic activity was inhibited in the presence of 2,3-bisphosphoglycerate and p-hydroxymercuribenzoate. Enzymic activity was inhibited in the presence of 2,3-bisphosphoglycerate and p-hydroxymercuribenzoate. The presence of an SH2 domain and proline-rich sequence motifs within hp51CN suggests that this 5-phosphatase interacts with various **proteins** in signal transduction.

L8 ANSWER 16 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1996:536750 CAPLUS
DOCUMENT NUMBER: 125:239226
TITLE: Neurotrophin-3 and brain-derived neurotrophic factor activate multiple signal transduction events but are not survival factors for hippocampal pyramidal neurons
AUTHOR(S): Marsh, H. Nicholas; Palfrey, H. Clive
CORPORATE SOURCE: Dep. Pharmacol. Physiol. Sci., Univ. Chicago, Chicago, IL, 60637, USA
SOURCE: J. Neurochem. (1996), 67(3), 952-963
CODEN: JONRA9; ISSN: 0022-3042
Searcher : Shears 308-4994

DOCUMENT TYPE: Journal
LANGUAGE: English

AB Expression of the neurotrophin-3 (NT-3) receptor (TrkC) and the effects of NT-3 on signal transduction were investigated in highly enriched populations of embryonic rat hippocampal pyramidal neurons grown in bilaminar cultures. **PCR** anal. revealed that the predominant trkC isoform is K1, which lacks an insert in the kinase domain. Polyclonal TrkC-specific antibodies stained >90% of the neurons and revealed a single .apprx.145-kDa **protein** in immunoblots of exts. from adult hippocampus and pyramidal neuron cultures. Addn. of NT-3 (50 ng/mL) to these cultures induced the tyrosine phosphorylation of TrkC but not TrkB, as detd. by anti-phosphotyrosine staining of immunoppts.; thus, all the effects of NT-3 are mediated through TrkC. NT-3 also increased the tyrosine phosphorylation of 42-, 44-, 49-, 55-, 95-, and 145-kDa **proteins**; the pattern induced by brain-derived neurotrophic factor (BDNF) was similar but not identical to that induced by NT-3, suggesting that subtle differences may exist in signaling by TrkB and TrkC receptors. Immunopptn. of p21ras from 32P-prelabeled cells showed that NT-3 increased the level of the GTP-bound form of the **protein** threefold over the control within 5 min. Mitogen-activated **protein** (MAP) kinase activity was maximally elevated by NT-3 within 2 min and then returned slowly toward baseline over the next 60 min. Tyrosine phosphorylation of phospholipase C-.gamma. increased rapidly after NT-3, suggesting that this enzyme becomes activated. Consistent with this, the neurotrophin rapidly increased **protein** kinase C activity as well as intracellular Ca2+ levels. The effects of both NT-3 and BDNF on Ca2+ levels were attenuated in Ca2+-free medium, suggesting that both neurotrophins increase Ca2+ flux across the plasma membrane as well as release from internal stores. NT-3 also increased c-Fos expression in >80% of the cells; the effect peaked at 30 min and declined to baseline by 120 min. Despite the activation of ras-MAP kinase and **phosphoinositide** signaling pathways, neither NT-3 nor BDNF alone or in combination could sustain hippocampal pyramidal neurons deprived of glial support. The authors conclude that in this system NT-3 and BDNF do not appear to be acting as classical "neurotrophic" factors and that activation of the MAP kinase pathway is insufficient for the promotion of neuronal survival.

L8 ANSWER 17 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1996:347029 CAPLUS
DOCUMENT NUMBER: 125:52266
TITLE: CpK is a novel class of Drosophila PtdIns 3-kinase containing a C2 domain
AUTHOR(S): Molz, Lisa; Chen, Yen-Wen; Hirano, Michiko; Williams, Lewis T.
CORPORATE SOURCE: Cardiovascular Res. Inst. Daiichi Res. Cent.,

Univ. California, San Francisco, CA, 94143, USA
SOURCE: J. Biol. Chem. (1996), 271(23), 13892-13899
CODEN: JBCHA3; ISSN: 0021-9258
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The authors report the identification of a novel class of phosphatidylinositol (PtdIns) 3-kinases whose members contain C-terminal C2 domains. The authors have isolated Drosophila and murine **genes** (termed cpk and cpk-m resp.) by **polymerase** chain **reaction** amplification of cDNA libraries with degenerate primers corresponding to conserved regions of PtdIns kinases. The amino acid sequences of Cpk and Cpk-m are most similar to that of p110, a family of PtdIns 3-kinases that mediates the responses of cells to mitogenic stimuli. The Cpk and Cpk-m sequences are similar to a large, central region of p110, but differ from p110 at their N and C termini. The N termini of the Cpk **proteins** do not contain any recognizable **protein** motif, while the C termini contain "C2 domains," a feature unique among PtdIns kinases. Cpk has an intrinsic PtdIns kinase activity and can phosphorylate PtdIns and PtdIns-4-P, but not PtdIns(4,5)P2, at the D3 position of the inositol ring. Cpk is the first PtdIns 3-kinase identified with this particular substrate specificity. The authors have identified two potential Cpk-binding **proteins**, p90 and p190, and have detd. that both Cpk and p190 may be tyrosine phosphorylated. This finding suggests that Cpk function may be regulated by tyrosine kinases.

IT **115926-52-8, Phosphatidylinositol 3-kinase**
RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
(isoenzyme; sequence of **gene** cpk encoding a novel class of Drosophila and mouse PtdIns 3-kinase contg. a C2 domain)

L8 ANSWER 18 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1996:80759 CAPLUS
DOCUMENT NUMBER: 124:168636
TITLE: A presynaptic inositol-5-phosphatase
AUTHOR(S): McPherson, Peter S.; Garcia, Elizabeth P.; Slepnev, Vladimir I.; David, Carol; Zhang, Xiaomei; Grabs, Detlev; Sossin, Wayne S.; Bauerfeind, Rudolf; Nemoto, Yasuo; De Camilli, Pietro
CORPORATE SOURCE: Dep. Cell Biology, Yale Univ. School of Medicine, New Haven, CT, 06510, USA
SOURCE: Nature (London) (1996), 379(6563), 353-57
CODEN: NATUAS; ISSN: 0028-0836
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Synaptojanin is a nerve terminal **protein** of relative mol.

mass 145,000 which appears to participate with dynamin in synaptic vesicle recycling. **Peptide** sequences from purified synaptojanin were used to design oligonucleotides for **PCR** amplification and characterization of a full-length cDNA from rat brain. The open reading frame encodes a putative **protein** of 1292 amino acids with a predicted isoelec. point of 6.96 , a predicted relative mol. mass of 142,797, and no putative transmembrane regions. A second in-frame ORF for 266 amino acids was obsd. after the UAA stop codon for synaptojanin. Neonate brain and undifferentiated PC-12 cells contain, in addn. to the 145-kDa band, a 170-kDa band which represents a longer form of synaptojanin contg. both ORF1 and ORF2. The central region of synaptojanin defines it as a member of the inositol-5-phosphatase family, which includes the product of the **gene** that is defective in the oculocerebrorenal syndrome of Lowe. Synaptojanin has 5-phosphatase activity and its N-terminal domain is homologous with the yeast **protein** Sac1 (Rsd1), which is genetically implicated in phospholipid metab. and in the function of the actin cytoskeleton. The C-terminus, which is of different lengths in adult and developing neurons owing to the alternative use of 2 termination sites, is proline-rich, consistent with the reported interaction of synaptojanin with the SH3 domains of Grb2. Synaptojanin is the only other major brain **protein** besides dynamin that binds the SH3 domain of amphiphysin, a presynaptic **protein** with a putative function in endophysin, a presynaptic **protein** with a putative function in endocytosis. The results suggest a link between **phosphoinositide** metab. and synaptic vesicle recycling.

L8 ANSWER 19 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1996:29607 CAPLUS
DOCUMENT NUMBER: 124:110218
TITLE: Cloning of the rat ErbB3 cDNA and characterization of the recombinant **protein**
AUTHOR(S): Hellyer, Nathan J.; Kim, Hong-Hee; Greaves, Charles H.; Sierke, Susan L.; Koland, John G.
CORPORATE SOURCE: Dep. of Pharmacology, Univ. of Iowa Coll. of Medicine, Iowa City, IA, 52242-1109, USA
SOURCE: Gene (1995), 165(2), 279-84
CODEN: GENED6; ISSN: 0378-1119
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Three cDNA fragments that encoded all but the extreme N terminus of the rat ErbB3 **protein** were cloned by low-stringency screening of a rat liver cDNA library with a human ERBB3 probe. The remaining 5'-end of the cDNA was generated by a reverse transcription-**polymerase** chain **reaction** method, and a single full-length rat ErbB3 cDNA was assembled. A comparison

of the deduced amino acid (aa) sequences of human and rat ErbB3 was made, and the effects of certain aa substitutions in the putative **protein** tyrosine kinase domain were considered. The rat ErbB3 cDNA was subsequently expressed in cultured NIH-3T3 mouse fibroblasts, in which a high level of approx. 180-kDa recombinant ErbB3 (re-ErbB3) was generated. The rat re-ErbB3 produced in transfected fibroblasts was responsive to the **polypeptide**, heregulin, a known ligand for ErbB3. Challenge of transfected fibroblasts with heregulin stimulated the phosphorylation of rat re-ErbB3 on Tyr residues and promoted its assocn. with the p85 subunit of **phosphatidylinositol 3-kinase** . Together, these results indicate that a fully functional rat ErbB3 cDNA has been isolated, and that fibroblast cells expressing this cDNA will be suitable for investigations of the signal transduction mechanism of ErbB3.

L8 ANSWER 20 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1996:10328 CAPLUS
DOCUMENT NUMBER: 124:80611
TITLE: A family of **phosphoinositide** 3-kinases in Drosophila identifies a new mediator of signal transduction
AUTHOR(S): MacDougall, Lindsay K.; Domin, Jan; Waterfield, Michael D.
CORPORATE SOURCE: Ludwig Inst. for Cancer Research, Univ. College Branch, London, W1P 8BT, UK
SOURCE: Curr. Biol. (1995), 5(12), 1404-15
CODEN: CUBLE2; ISSN: 0960-9822
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Mammalian **phosphoinositide** 3-kinases (PI 3-kinases) are involved in receptor-mediated signal transduction and have been implicated in processes such as transformation and mitogenesis through their role in elevating cellular phosphatidylinositol (3,4,5)-triphosphate. Addnl., a PI 3-kinase activity which generates phosphatidylinositol 3-phosphate has been shown to be required for **protein** trafficking in yeast. The authors have identified a family of three distinct PI 3-kinases in Drosophila, using an approach based on the **polymerase** chain **reaction** to amplify a region corresponding to the conserved catalytic domain of PI 3-kinases. One of these family members, PI3K 92D, is closely related to the prototypical PI 3-kinase, p110.alpha.; PI3K 59F is homologous to Vps34p, whereas the third, PI3K 68D, is a novel PI 3-kinase which is widely expressed throughout the Drosophila life cycle. The PI3K 68D cDNA encodes a **protein** of 210 kDa, which lacks sequences implicated in linking p110 PI 3-kinases to p85 adaptor **proteins**, but contains an amino-terminal proline-rich sequence, which could bind to SH3 domains, and a carboxy-terminal C2 domain. Biochem. analyses

demonstrate that PI3K 68D has a novel substrate specificity in vitro, restricted to phosphatidylinositol and phosphatidylinositol 4-phosphate, and is unable to phosphorylate phosphatidylinositol (4,5)-bisphosphate, the implied in vivo substrate for p110. A family of PI 3-kinases in Drosophila, including a novel class represented by PI3K 68D, is described PI3K 68D has the potential to bind to signaling mols. contg. SH3 domains, lacks p85-adaptor-binding sequences, has a Ca2+-independent phospholipid-binding domain and displays a restricted in vitro substrate specificity, so it could define a novel signal transduction pathway.

IT **115926-52-8, Phosphoinositide** 3-kinase
RL: BAC (Biological activity or effector, except adverse); BPR (Biological process); PRP (Properties); BIOL (Biological study); PROC (Process)
(amino acid sequence and novel substrate specificity in vitro; family of **phosphoinositide** 3-kinases in Drosophila identifies new mediator of signal transduction)

L8 ANSWER 21 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1995:1001786 CAPLUS
DOCUMENT NUMBER: 124:78206
TITLE: Identification of two isoforms of mouse neuropeptide Y-Y1 receptor generated by alternative splicing. Isolation, genomic structure, and functional expression of the receptors
AUTHOR(S): Nakamura, Motonao; Sakanaka, Chie; Aoki, Yoshiko; Ogasawara, Hiroyuki; Tsuji, Takashi; Kodama, Hisashi; Matsumoto, Takashi; Shimizu, Takao; Noma, Masana
CORPORATE SOURCE: Life Science Res. Laboratory, Japan Tobacco Inc., Yokohama, 227, Japan
SOURCE: J. Biol. Chem. (1995), 270(50), 30102-10
CODEN: JBCHA3; ISSN: 0021-9258
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Two cDNA clones homologous with human neuropeptide (NP) Y-Y1 receptor have been isolated from a mouse bone marrow cDNA library. One was thought to be the cognate of the human NPY-Y1 receptor, termed Y1.alpha. receptor, and the other form, termed Y1.beta. receptor, differed from the Y1.alpha. receptor in the seventh transmembrane domain and C-terminal tail. Anal. of the mouse genomic **DNA** showed that both receptors originated from a single **gene**. The different **peptide** sequences of the Y1.beta. receptor were encoded by sep. exons, hence, these receptors were generated by differential **RNA** splicing. High affinity binding of [125I]NPY to each receptor expressed in Chinese hamster ovary (CHO) cells and sequestration of [125I]NPY

after binding to cells and sequestration of [125I]NPY after binding to each receptor were obsd. In the CHO cells expressing the Y1.alpha. receptor, intracellular Ca2+ increase, inhibition of forskolin-induced cAMP accumulation, and mitogen-activated **protein** kinase (MAPK) activation were obsd. by stimulation of NPY, and these responses were abolished by pretreatment with pertussis toxin. Since wortmannin completely inhibited NPY-elicited MAPK activation, we speculate that wortmannin-sensitive signaling mol.(s) such as **phosphoinositide** 3-kinase may lie between pertussis toxin-sensitive G-**protein** and MAPK. In contrast, these intracellular signals were not detected in CHO cells expressing the Y1.beta. receptor. Northern blots and reverse transcriptase-**polymerase** chain **reaction** analyses indicated that the Y1.alpha. receptor was highly expressed in the brain, heart, kidney, spleen, skeletal muscle, and lung, whereas the Y1.beta. receptor mRNA was not detected in these tissues. However, the Y1.beta. receptor was expressed in mouse embryonic development stage (7 and 11 days), bone marrow cells and several hematopoietic cell lines. These results suggest that the Y1.beta. receptor is an embryonic and a bone marrow form of the NPY-Y1 receptor, which decreases in the expression during development and differentiation.

IT **115926-52-8, Phosphoinositide** 3-kinase
RL: BAC (Biological activity or effector, except adverse); BIOL (Biological study)
(since wortmannin completely inhibited mouse neuropeptide Y-Y1-elicited mitogen-activated **protein** kinase, wortmannin-sensitive signaling mol.(s) as **phosphoinositide** 3-kinase may lie between pertussis toxin-sensitive G-**protein** and MAPK)

L8 ANSWER 22 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1995:962541 CAPLUS
DOCUMENT NUMBER: 124:107472
TITLE: Molecular cloning and expression of multiple isoforms of human prostaglandin E receptor EP3 subtype generated by alternative messenger **RNA** splicing: multiple second messenger systems and tissue-specific distributions
AUTHOR(S): Kotani, Masato; Tanaka, Issei; Ogawa, Yoshihiro; Usui, Takeshi; Mori, Kiyoshi; Ichikawa, Atsushi; Narumiya, Shuh; Yoshimi, Teruya; Nakao, Kazuwa
CORPORATE SOURCE: Dep. Med., Kyoto Univ., Kyoto, Japan
SOURCE: Mol. Pharmacol. (1995), 48(5), 869-79
CODEN: MOPMA3; ISSN: 0026-895X
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Five distinct cDNA clones encoding 4 different isoforms of human prostaglandin (PG) E receptor EP3 subtype were isolated from a human kidney cDNA library. Two cDNA clones differed only in their

3'-untranslated regions. The 4 isoforms, tentatively named EP3-I, EP3-II, EP3-III, and EP3-IV, which were generated by alternative mRNA splicing, had identical amino acid sequences except for their different C-terminal tails. Transfection expts. revealed that all the 4 isoforms show high binding affinities to PGE2, PGE1, and M&b28767, an EP3-specific agonist, whereas their downstream signaling pathways are divergent. M&B28767 increased cAMP concns. in cells expressing EP3-II and EP3-IV, whereas it inhibited forskolin-induced cAMP accumulations in cells expressing all EP3 isoforms. M&B28767 also stimulated **phosphoinositide** turnover in cells expressing EP3-I and EP3-II. Northern blot anal. revealed that the EP3 **gene** is expressed in a wide variety of human tissues. The human EP3 mRNA was present most abundantly in the kidney, pancreas, and uterus. A substantial expression was also detected in the heart, liver, skeletal muscle, small intestine, colon, prostate, ovary, and testis. Furthermore, reverse transcription-**polymerase** chain **reaction** anal. demonstrated tissue-specific expressions of the 5 different EP3 mRNA species. The present study suggests the presence of the multiple systems of PGE2/EP3 isoforms and leads to the better understanding of its physiol. and pathophysiol. implications in humans.

L8 ANSWER 23 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1995:808970 CAPLUS
DOCUMENT NUMBER: 123:333394
TITLE: cDNA encoding a functional feline liver/bone/kidney-type alkaline phosphatase
AUTHOR(S): Ghosh, Ananta K.; Mullins, James I.
CORPORATE SOURCE: Dep. Microbiol. Immunol., Stanford Univ. Sch. Med., Stanford, CA, 94305-5402, USA
SOURCE: Arch. Biochem. Biophys. (1995), 322(1), 240-49
CODEN: ABBIA4; ISSN: 0003-9861
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Feline alk. phosphatase (FeALP) was copurified with the putative 70-kDa feline leukemia virus subgroup-A (FeLV-A) receptor **protein** from feline T-lymphocyte cells (FeT) by two-dimensional gel electrophoresis. The sequence of the N-terminal 17 amino acids and five other internal tryptic **peptides** revealed that it is homologous to the liver/bone/kidney (L/B/K)-type alk. phosphatase of other mammalian species. Corresponding oligonucleotides were synthesized and used for amplification of a 1.2-kb segment of the FeALP **gene** by **polymerase** chain **reaction**, using phage **DNA** from a FeT cell cDNA library as template. The 1.2-kb FeALP **gene** fragment generated was then used as a probe to isolate a 2127-bp L/B/K-type FeALP cDNA clone from the same library harboring a large, intact open reading frame. This cDNA possessed an open reading frame encoding a 524-amino-acid **protein** including a putative

signal **peptide** of 17 amino acids followed by 14-amino-acid residues identical to the N-terminal sequence detd. from the purified **protein**. Sequences closely related to five tryptic **peptides** from the purified **protein** were also contained within the cDNA-encoded **protein**. Homol. with the human, bovine, rat and mouse L/B/K-type ALP was found to be 88-90% at both the nucleotide and the amino acid levels. The cDNA was transferred into a eukaryotic expression vector and expressed following transfection into murine and mink lung fibroblast cell lines. High levels of enzymically active ALP were detected, along with a 70-kDa **protein** reactive in immunoblot assay using a polyclonal antibody against the original crude FeALP prepn. FeALP was specifically released from intact cells by treatment with **phosphoinositol**-specific phospholipase-C. By Northern blot anal., only one species of mRNA was detected using a 32P-labeled cDNA probe. These results indicate that the 2127-bp cDNA encodes a functional feline L/B/K-type ALP expressed on cell surfaces via phosphatidylinositol-glycan linkage. Despite electrophoretic comigration in two dimensions and following deglycosylation in a third dimension, FeALP failed to function as an FeLV receptor since its expression failed to provide for attachment or entry of virus into cells.

L8 ANSWER 24 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1995:361281 CAPLUS
DOCUMENT NUMBER: 122:307757
TITLE: Molecular cloning of the plc1+ **gene** of Schizosaccharomyces pombe, which encodes a putative **phosphoinositide**-specific phospholipase C
AUTHOR(S): Andoh, Tomoko; Yoko-O, Takehiko; Matsui, Yasushi; Toh-E, Akio
CORPORATE SOURCE: Grad. Sch. Sci., Univ. Tokyo, Tokyo, 113, Japan
SOURCE: Yeast (1995), 11(2), 179-85
CODEN: YESTE3; ISSN: 0749-503X
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Exploiting the **polymerase** chain **reaction**, the authors have isolated a **gene** that endoes a putative **phosphoinositide**-specific phospholipase C (PLC) of the fission yeast S. pombe. Inspection of the nucleotide sequence of the **gene** revealed an open reading frame that can encode a **polypeptide** of 899 amino acid residues with a calcd. mol. mass of 102 kDa. This putative **polypeptide** contains both the X and Y regions that are conserved among 3 classes of mammalian PLC, and also contains a presumptive Ca2+-binding site (an E-F hand motif). The structure of the putative **protein** is most similar to that of the .delta. class of PLC isoenzymes. To investigate the role of this structure of the putative

protein is most similar to that of the .delta. class of PLC isoenzymes. To investigate the role of this **gene**, designated plc1+, **gene** disruption was carried out by interrupting the coding region with the ura4+ marker. Growth of plc1 cells was temp.-sensitive in rich medium, and cells could not grow in synthetic medium. Expression of the PLC1 **gene** of Saccharomyces cerevisiae suppressed the growth defect phenotype of plc1- cells, a strong suggestion that the plc1+ **gene** encodes PLC. The PLC1 sequence appears in the public data libraries, DDBJ GenBank, EMBL under the following Accession No.: D38309.

L8 ANSWER 25 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1995:303601 CAPLUS
DOCUMENT NUMBER: 122:206627
TITLE: Molecular cloning, cDNA sequence, and chromosomal localization of the human **phosphatidylinositol 3-kinase** p110.alpha. (PIK3CA) **gene**
AUTHOR(S): Volinia, Stefano; Hiles, Ian; Ormondroyd, Elizabeth; Nizetic, Dean; Antonacci, Rachele; Rocchi, Mariano; Waterfield, Michael D.
CORPORATE SOURCE: Ludwig Inst. Cancer Res., London, W1P 8BT, UK
SOURCE: Genomics (1994), 24(3), 472-7
CODEN: GNMCEP; ISSN: 0888-7543
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Phosphatidylinositol (PI) 3-kinase is a heterodimeric enzyme comprising a 110-kDa catalytic subunit and an 85-kDa regulatory subunit that binds to tyrosine phosphopeptide sites linked directly or indirectly to receptors serving diverse signal functions. Knowledge of the structure and function of PI 3-kinase was greatly advanced by the purifn., cDNA cloning, and subsequent expression of the bovine enzyme. Here, the cloning of the cDNA for the human p110.alpha. subunit of **PI3-kinase** (PIK3CA), encoding a **protein** 99% identical to the bovine p110, and of its **gene** in YAC is described. The chromosomal localization of the **gene** for PIK3CA is shown to be at 3q21-qter as detd. using somatic cell hybrids. In situ hybridization performed using Alu-**PCR** from the YAC **DNA** located the **gene** in 3q26.3.

IT **115926-52-8**, **Phosphatidylinositol 3-kinase**
RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
(mol. cloning, cDNA sequence, and chromosomal localization of human **phosphatidylinositol 3-kinase** p110.alpha. (PIK3CA) **gene**)

L8 ANSWER 26 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1994:573797 CAPLUS
DOCUMENT NUMBER: 121:173797
TITLE: Cloning and expression of a novel, highly truncated **phosphoinositide**-specific phospholipase C cDNA from embryos of the brine shrimp, Artemia
AUTHOR(S): Su, Xilin; Chen, Fengling; Hokin, Lowell E.
CORPORATE SOURCE: Med. Sch., Univ. Wisconsin, Madison, WI, 53706, USA
SOURCE: J. Biol. Chem. (1994), 269(17), 12925-31
CODEN: JBCHA3; ISSN: 0021-9258
DOCUMENT TYPE: Journal
LANGUAGE: English
AB A novel, truncated form of a cDNA encoding Artemia **phosphoinositide**-specific phospholipase C (PLC), designated PLC-.beta.x, was isolated from a brine shrimp cDNA library. The full-length cDNA is of the .beta.-type, it is 2855 base pairs long, and it contains an open reading frame encoding 489 amino acids. The deduced amino acid sequence of PLC-.beta.x cDNA shows novel, features. It lacks several hundred amino acids at the 5' end, as compared to PLC-.beta.s in the higher species. It contains conserved domains X and Y, but domain X is highly truncated at the 5' end (only 14-25 conserved amino acids as compared to about 150 amino acids in the higher eukaryotic organisms). Northern blot hybridization showed that the PLC-.beta.x cDNA corresponds to a 4.4-kilobase mRNA. Northern blot hybridization with a cDNA probe from the 5' end and **PCR** performed upstream from domain Y showed that PLC-.beta.x is not a cloning artifact due to fusion of unrelated clone into the coding region of the PLC-.beta. homolog. A functional PLC and new **protein** bands on SDS-PAGE were obsd. after subcloning full-length PLC-.beta.x cDNA, as well as a fragment contg. the conserved regions, into expression plasmids vectors and transfecting into Escherichia coli. 1 MM lithium markedly stimulated expression in E. coli.

L8 ANSWER 27 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1994:505475 CAPLUS
DOCUMENT NUMBER: 121:105475
TITLE: Studies on biological implication of gonadotropin-releasing hormone (Gn-RH) and its receptor in human ovarian carcinoma
AUTHOR(S): Ohno, Tsukasa; Imai, Atsushi; Tamaya, Teruhiko
CORPORATE SOURCE: Sch. Med., Gifu Univ., Gifu, 500, Japan
SOURCE: Gifu Daigaku Igakubu Kiyo (1994), 42(2), 174-85
CODEN: GDIKAN; ISSN: 0072-4521
DOCUMENT TYPE: Journal
LANGUAGE: Japanese
AB This study was designed to investigate the biol. implications of

gonadotropin-releasing hormone (Gn-RH) and its receptor in human ovarian carcinoma, in order to know the possible clin. use of Gn-RHA analogs in the treatment of ovarian carcinoma. Gn-RH was detd. by a RIA and bioassay. Gn-RH mRNA was detd. by reverse transcription (RT)-**PCR** using oligonucleotide primers synthesized according to the published Gn-RH **gene** sequence. Gn-RH was shown to be present in exts. of ovarian mucinous cystadenocarcinoma samples and ovarian adenocarcinoma cell line SK-OV-3, but not in normal ovary and placenta. Two of two ext. samples from individual cases of mucinous cystadenocarcinoma evoked dose-dependent **phosphoinositide** breakdown in rat granulosa cells, similar to that caused by authentic Gn-RH. Gn-RH mRNA was detected in 2 of 2 mucinous cystadenocarcinoma specimens, 1 of 1 serous cystadenocarcinoma, and SK-OV-3 cells, but not in mucinous cystadenoma, the normal ovary or placenta. Gn-RH receptor was detd. by the presence of [3H]Gn-RH binding sites and its mRNA by the RT-**PCR** method. The transmembrane signaling events interacting with Gn-RH receptor were also established in **phosphoinositide** turnover. Specific Gn-RH binding sites were shown to be present in the plasma membrane isolated from 2 ovarian mucinous cystadenocarcinoma samples (kd = 8.0 nM, Bmax = 0.1-0.2 pmol/mg **protein**). Gn-RH receptor mRNA was detected in 2 of 2 mucinous cystadenocarcinoma specimens, 1 of 1 serous cystadenocarcinoma, and SK-OV-3 cells, but not in female white blood cells. In mucinous cystadenocarcinoma, incubation of plasma membranes isolated from [3H]inositol-labeled specimens with Gn-RH resulted in the rapid prodn. of inositol phosphates. Chronic exposure to Gn-RH analog buserelin attenuated the sensitivity of inositol phosphates formation to Gn-RH in the membrane event. The demonstrations of Gn-RH and its receptor raise the possibility that Gn-RH may play an autocrine regulatory role in the growth of ovarian carcinoma. The relatively high dose of Gn-RH analog could induce down-regulation of Gn-RH receptor and/or transmembrane signaling with a consequent suppression of the tumor growth.

L8 ANSWER 28 OF 32 CAPLUS COPYRIGHT 1999 ACS

ACCESSION NUMBER: 1994:188437 CAPLUS

DOCUMENT NUMBER: 120:188437

TITLE: Expression of three alternative acetylcholinesterase messenger **RNAs** in human tumor cell lines of different tissue origins

AUTHOR(S): Karpel, R.; Ben Aziz-Aloya, R.; Sternfeld, M.; Ehrlich, G.; Ginzberg, D.; Tarroni, P.; Clementi, F.; Zakut, H.; Soreq, H.

CORPORATE SOURCE: Life Sci. Inst., Hebrew Univ., Jerusalem, 91904, Israel

SOURCE: Exp. Cell Res. (1994), 210(2), 268-77
CODEN: ECREAL; ISSN: 0014-4827

DOCUMENT TYPE: Journal
LANGUAGE: English

AB To study the mol. mechanisms underlying the intensive expression of acetylcholinesterase (ACHE) in different tumor types, the authors characterized levels and compn. of its mRNA (mRNA) sequences in heterologous tumor cell lines, primary tumor biopsies, and normal fetal and adult tissues and detd. their exon-intron origin within the corresponding ACHE **gene**. Reverse transcription followed by **polymerase** chain **reaction** (RT-**PCR**) revealed three alternatively spliced ACHE mRNAs in NT2/D1 teratocarcinoma, NCI-N-592 small cell lung carcinoma, TE671 medulloblastoma, K-562 erythroleukemia, and 293 transformed embryonal kidney cells. The three ACHE mRNAs include the principal species expressed in brain and muscle and two addnl. transcripts contg. insertions of 751 or 829 residues downstream from the exon 4 domain. The inserted region, which represents an intron in brain and muscle, is expressed in the tumor cell lines either as a "readthrough" form or with 78 residues deleted from its 5' end. A major band of 2.5 kb was labeled with ACHE cDNA in poly(A)+ **RNA** blots from medulloblastoma cells or brain tissue, whereas a **PCR**-amplified probe from the inserted domain labeled a 3.4-kb band but not the 2.5-kb band in poly(A)+ **RNA** from small cell lung carcinoma. The ACHE mRNAs including the alternative insertions were found only in cell lines with levels of the principal ACHE mRNA species equal to or higher than those in brain (1-10 mols./cell), detd. by following the kinetics of mRNA **PCR** amplification. Genomic **DNA** sequencing revealed that the inserted domains in the ACHE mRNAs expressed in the tumor cell lines encode C-terminal **peptides** of 40 and 14 residues. These include a free cysteine, terminate with the consensus HG element, and continue by a 29-residue-long C-terminal hydrophobic cleavable **peptide**, properties characteristic of precursors to **phosphoinositide** (PI)-linked **proteins**. In extension of the reported expression of PI-linked ACHE in hemopoietic cells including K-562, the authors' findings demonstrate the existence of ACHE mRNAs with the potential to encode one hydrophilic and two PI-linked forms of ACHE in tumor cells from both hemopoietic and nonhemopoietic origins.

L8 ANSWER 29 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1994:100407 CAPLUS
DOCUMENT NUMBER: 120:100407
TITLE: Phosphatidylinositol 4-kinase: **gene** structure and requirement for yeast cell viability
AUTHOR(S): Flanagan, Catherine A.; Schnieders, Elisabeth A.; Emerick, Anne W.; Kunisawa, Riyo; Admon, Arie; Thorner, Jeremy

CORPORATE SOURCE: Dep. Mol. Cell Biol., Univ. California, Berkeley, CA, 94720, USA
SOURCE: Science (Washington, D. C., 1883-) (1993), 262(5138), 1444-8
CODEN: SCIEAS; ISSN: 0036-8075
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Phosphatidylinositol (Ptdlns) 4-kinase catalyzes the first step in the biosynthesis of Ptdlns-4,5-bisphosphate (Ptdlns[4,5]P2). Hydrolysis of Ptdlns[4,5]P2 in response to extracellular stimuli is thought to initiate intracellular signaling cascades that modulate cell proliferation and differentiation. The PIK1 **gene** encoding a Ptdlns 4-kinase from the yeast Saccharomyces cerevisiae was isolated by **polymerase** chain **reaction** (**PCR**) with oligonucleotides based on the sequence of **peptides** derived from the purified enzyme. The sequence of the PIK1 **gene** product bears similarities to that of Ptdlns 3-kinases from mammals (p110) and yeast (Vps34p). Expression of PIK1 from a multicopy plasmid elevated Ptdlns 4-kinase activity and enhanced the response to mating pheromone. A pik1 null mutant was inviable, indicating that Ptdlns4P and presumably Ptdlns[4,5]P2 are indispensable phospholipids.

L8 ANSWER 30 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1994:28447 CAPLUS
DOCUMENT NUMBER: 120:28447
TITLE: Identification of mutations in the coding sequence of the proto-oncogene c-kit in a human mast cell leukemia cell line causing ligand-independent activation of c-kit product
AUTHOR(S): Furitsu, Takuma; Tsujimura, Tohru; Tono, Toshiharu; Ikeda, Hirokazu; Kitayama, Hitoshi; Koshimizu, Uichi; Sugahara, Hiroyuki; Butterfield, Joseph H.; Ashman, Leonie K.; et al.
CORPORATE SOURCE: Med. Sch., Osaka Univ., Suita, 565, Japan
SOURCE: J. Clin. Invest. (1993), 92(4), 1736-44
CODEN: JCINAO; ISSN: 0021-9738
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The c-kit proto-oncogene encodes a receptor tyrosine kinase. Binding of c-kit ligand, stem cell factor (SCF), to c-kit receptor (c-kitR) is known to activate c-kitR tyrosine kinase, thereby leading to autophosphorylation of c-kitR on tyrosine and to assocn. of c-kitR with substrates such as **phosphatidylinositol 3-kinase** (PI3K). In a human mast cell leukemia cell line HMC-I, c-kitR was found to be constitutively phosphorylated on tyrosine, activated, and assocd. with PI3K without the addn. of SCF. The expression of SCF mRNA transcript in HMC-1

cells was not detectable by means of **PCR** after reverse transcription (RT-**PCR**) anal., suggesting that the constitutive activation of c-kitR was ligand independent. Sequencing of whole coding region of c-kit cDNA revealed that c-kit **genes** of HMC-1 cells were composed of a normal, wild-type allele and a mutant allele with two point mutations resulting in intracellular amino acid substitutions of Gly-560 for Val and Val-816 for Asp. Amino acid sequences in the regions of the two mutations are completely conserved in all of mouse, rat, and human c-kit. In order to det. the causal role of these mutations in the constitutive activation, murine c-kit mutants encoding Gly-559 and/or Val-814, corresponding to human Gly-560 and/or Val-816, were constructed by site-directed mutagenesis and expressed in a human embryonic kidney cell line, 293T cells. In the transfected cells, both c-kitR (Gly-559, Val-814) and c-kitR (Val-814) were abundantly phosphorylated on tyrosine and activated in immune complex kinase reaction in the absence of SCF, whereas tyrosine phosphorylation and activation of c-kitR (Gly-559) or wild-type c-kitR was modest or little, resp. These results suggest that conversion of Asp-816 to Val in human c-kitR may be an activating mutation and responsible for the constitutive activation of c-kitR in HMC-1 cells.

L8 ANSWER 31 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1993:248877 CAPLUS
DOCUMENT NUMBER: 118:248877
TITLE: The putative **phosphoinositide**-specific phospholipase C **gene**, PLC1, of the yeast Saccharomyces cerevisiae is important for cell growth
AUTHOR(S): Yokoo, Takehiko; Matsui, Yasushi; Yagisawa, Hitoshi; Nojima, Hiroshi; Uno, Isao; Tohe, Akio
CORPORATE SOURCE: Fac. Sci., Univ. Tokyo, Tokyo, 113, Japan
SOURCE: Proc. Natl. Acad. Sci. U. S. A. (1993), 90(5), 1804-8
CODEN: PNASA6; ISSN: 0027-8424
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Using the **polymerase** chain **reaction** technique, a **gene** that encodes a putative **phosphoinositide** -specific phospholipase C (PLC) in the yeast S. cerevisiae was isolated. The nucleotide sequence indicates that the **gene** encodes a **polypeptide** of 869 amino acid residues with a calcd. mol. mass of 101 kDa. This **polypeptide** has both the X and Y regions conserved among mammalian PLC-.beta., -.gamma., and -.delta., and the structure is most similar to that of mammalian PLC-.delta.. This putative yeast PLC **gene** has been designated PLC1. Disruption of PLC1 results in slow growth or lethality for cells, depending on their genetic background and the medium, indicating that PLC1 is important for cell growth.

Expression of rat PLC-.delta.1 cDNA suppressed the growth defect of plc1 disruptants, strongly suggesting that PLC1 encodes PLC.

L8 ANSWER 32 OF 32 CAPLUS COPYRIGHT 1999 ACS
ACCESSION NUMBER: 1993:208397 CAPLUS
DOCUMENT NUMBER: 118:208397
TITLE: Molecular cloning and expression of a **phosphoinositide**-specific phospholipase C of Dictyostelium discoideum
AUTHOR(S): Drayer, A. Lyndsay; Van Haastert, Peter J. M.
CORPORATE SOURCE: Dep. Biochem., Univ. Groningen, Groningen, 9747 AG, Neth.
SOURCE: J. Biol. Chem. (1992), 267(26), 18387-92
CODEN: JBCHA3; ISSN: 0021-9258
DOCUMENT TYPE: Journal
LANGUAGE: English
AB A no. of **phosphoinositide**-specific phospholipases C (PLC) of different species have recently been cloned. The predicted amino acid sequences of these isoforms contain two highly conserved domains. A PLC **gene** of Dictyostelium was identified using the **polymerase** chain **reaction**. Primers were designed coding for highly conserved amino acid regions located within one of the conserved domains of PLCs. Cloning and sequencing of the **polymerase** chain **reaction** product revealed one unique PLC-like sequence. This sequence was used to screen a library and isolate several overlapping cDNA clones. The complete cDNA was expressed in Dictyostelium cells resulting in increased basal levels of inositol 1,4,5-trisphosphate and enhanced PLC activity. The identified Dictyostellium PLC, DdPLC, encodes a **protein** with a calcd. mol. mass of 91 kDa. The deduced amino acid sequence contains the two conserved domains found in other PLC isoforms, sepd. by a short variable region. The C-terminal part of the **protein** shows strong homol. with the mammalian PLC-.delta. isoform. DdPLC is expressed at all stages of development, with an increase in transcription during starvation and in the culminating fruiting body.

=> d his l9- ful; d 1-59 ibib abs

(FILE 'MEDLINE, BIOSIS, EMBASE, LIFESCI, BIOTECHDS, WPIDS, CONFSCI, SCISEARCH, JICST-EPLUS, PROMT' ENTERED AT 14:23:40 ON 16 SEP 1999)
L9 169 SEA ABB=ON PLU=ON L8
L10 112 DUP REM L9 (57 DUPLICATES REMOVED)
L11 21778 SEA ABB=ON PLU=ON L2(L)(POLYPEPTIDE OR POLYPROTEIN OR PEPTIDE OR PROTEIN)
L12 287 SEA ABB=ON PLU=ON L11(L)(PCR OR POLYMERASE(1W) REACT?)
L13 75 SEA ABB=ON PLU=ON L12(L)(DNA OR DEOXYRIBONUCLEIC OR RNA OR RIBONUCLEIC OR NUCLEIC)
L14 48 SEA ABB=ON PLU=ON L13(L) GENE
Searcher : Shears 308-4994

L15 14 DUP REM L14 (34 DUPLICATES REMOVED)
L16 384 SEA ABB=ON PLU=ON L2(L)(PCR OR POLYMERASE(1W) REACT?)
L17 324 SEA ABB=ON PLU=ON L16 AND (POLYPEPTIDE OR POLYPROTEIN OR PEPTIDE OR PROTEIN)
L18 175 SEA ABB=ON PLU=ON L17 AND (DNA OR DEOXYRIBONUCLEIC OR RNA OR RIBONUCLEIC OR NUCLEIC)
L19 108 SEA ABB=ON PLU=ON L18 AND GENE
L20 108 SEA ABB=ON PLU=ON L14 OR L19
L21 59 DUP REM L20 (49 DUPLICATES REMOVED)

L21 ANSWER 1 OF 59 EMBASE COPYRIGHT 1999 ELSEVIER SCI. B.V.
ACCESSION NUMBER: 1999163702 EMBASE
TITLE: Characterization of a naturally occurring ErbB4 isoform that does not bind or activate phosphatidyl inositol 3-kinase.
AUTHOR: Elenius K.; Choi C.J.; Paul S.; Santiestevan E.; Nishi E.; Klagsbrun M.
CORPORATE SOURCE: M. Klagsbrun, Department of Surgery, Children's Hospital, Harvard Medical School, Boston, MA 02115, United States
SOURCE: Oncogene, (22 Apr 1999) 18/16 (2607-2615).
Refs: 55
ISSN: 0950-9232 CODEN: ONCNES
COUNTRY: United Kingdom
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Receptor tyrosine kinases regulate cell behavior by activating specific signal transduction cascades. Epidermal growth factor (EGF) receptor tyrosine kinases include ErbB1, ErbB2, ErbB3 and ErbB4. ErbB4 is a tyrosine kinase receptor that binds neuregulins (NRG) and several other EGF family members. Reverse transcriptase **polymerase** chain **reaction** (RT-**PCR**) analysis identified two isoforms of ErbB4 that differed in their cytoplasmic domain sequences. Specifically, RT-**PCR** using primers flanking the putative **phosphatidyl inositol 3-kinase** (PI3-K) binding site of ErbB4 generated two specific bands when human and mouse heart and kidney tissues were analysed. Cloning and sequencing of these RT-**PCR** products revealed that one of the ErbB4 isoforms (ErbB4 CYT-2) lacked a 16 amino acid sequence including a putative PI3-K binding site, that was present in the other isoform (ErbB4 CYT-1). RT-**PCR** analysis of mouse tissues suggested that the expression of ErbB4 CYT-1 and ErbB4 CYT-2 was tissue-specific. Heart, breast and abdominal aorta expressed predominantly ErbB4 CYT-1 whereas neural tissues and kidney expressed predominantly ErbB4 CYT-2. To ascertain whether the absence of the putative PI3-K binding site in ErbB4 CYT-2 also resulted in the loss of PI3-K

activity, NIH3T3 cell lines overexpressing ErbB4 CYT-1 or ErbB4 CYT-2 were produced. NRG-1 bound to and stimulated equivalent tyrosine phosphorylation of both isoforms. However, unlike ErbB4 CYT-1, the ErbB4 CYT-2 isoform was unable to bind the p85 subunit of PI3-K and to stimulate PI3-K activity in these cells. Furthermore, tyrosine phosphorylation of p85 or association of PI3-K activity with phosphotyrosine was not induced in NRG-1 treated cells expressing ErbB4 CYT-2, indicating that this isoform was incapable of activating PI3-K even indirectly. It was concluded that a novel naturally occurring ErbB4 isoform exists with a deletion of the cytoplasmic domain sequence required for the activation of the PI3-K intracellular signal transduction pathway and that this is the only PI3-K binding site in ErbB4.

L21 ANSWER 2 OF 59 MEDLINE
ACCESSION NUMBER: 1999355030 MEDLINE
DOCUMENT NUMBER: 99355030
TITLE: Repeated administration of dexamethasone increases phosphoinositide-specific phospholipase C activity and mRNA and **protein** expression of the phospholipase C beta 1 isozyme in rat brain.
AUTHOR: Dwivedi Y; Pandey G N
CORPORATE SOURCE: Psychiatric Institute, Department of Psychiatry, College of Medicine, University of Illinois at Chicago, 60612, USA.
CONTRACT NUMBER: RO 1-MH 56528 (NIMH)
SOURCE: JOURNAL OF NEUROCHEMISTRY, (1999 Aug) 73 (2) 780-90. Journal code: JAV. ISSN: 0022-3042.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199910
ENTRY WEEK: 19991003

AB Altered hypothalamic-pituitary-adrenal (HPA) function has been shown to be associated with changes in mood and behavior. The enzyme **phosphoinositide**-specific phospholipase C (PI-PLC), an important component of the PI signal transduction system, plays a major role in mediating various physiological functions. In the present study, we investigated the effects of a single dose and of repeated administration (0.5 or 1.0 mg/kg for 10 days) of dexamethasone (DEX), a synthetic glucocorticoid, on PI-PLC activity and on expression of PLC isozymes (beta1, delta1, and gamma1) in rat brain. Repeated administration of DEX (1.0 mg/kg) caused a significant increase in PI-PLC activity and in **protein** expression of the PLC beta1 isozyme in both membrane and cytosol fractions of cortex and hippocampus; however, the repeated administration of a smaller dose of DEX (0.5 mg/kg) caused these changes only in hippocampus but not in cortex. The increase in PLC

beta1 **protein** was associated with an increase in its mRNA level, as measured by competitive RT-**PCR**. A single administration of DEX (0.5 or 1.0 mg/kg) to rats had no significant effects on PI-PLC activity or on the **protein** expression of PLC isozymes. These results suggest that DEX up-regulates PI-PLC in rat brain, which presumably is due to a selective increase in expression of the PLC beta1 isozyme, and that these changes in PI-PLC may be related to HPA axis-mediated changes in mood and behavior.

L21 ANSWER 3 OF 59 MEDLINE DUPLICATE 1
ACCESSION NUMBER: 1999155281 MEDLINE
DOCUMENT NUMBER: 99155281
TITLE: Pituitary adenylate cyclase-activating **polypeptide** modulates gastric enterochromaffin-like cell proliferation in rats.
AUTHOR: Lauffer J M; Modlin I M; Hinoue T; Kidd M; Zhang T; Schmid S W; Tang L H
CORPORATE SOURCE: Gastric Pathobiology Research Group, Department of Surgery, Yale University School of Medicine and West Haven Veterans Administration Medical Center, New Haven, Connecticut, USA.
CONTRACT NUMBER: DK-48820 (NIDDK)
SOURCE: GASTROENTEROLOGY, (1999 Mar) 116 (3) 623-35. Journal code: FH3. ISSN: 0016-5085.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals; Cancer Journals
ENTRY MONTH: 199905
ENTRY WEEK: 19990504
AB BACKGROUND & AIMS: Gastric carcinoids (types I and II) involve the transformation of naive enterochromaffin-like (ECL) cells to the neoplastic state and are associated primarily with hypergastrinemia. In this study, we evaluated the effects of two related neuropeptides, pituitary adenylate cyclase-activating **polypeptide** (PACAP) and vasoactive intestinal **polypeptide** (VIP), on ECL cell proliferation and characterized the receptor subtype(s) and signal transduction pathways that mediate this effect. METHODS: Purified rat ECL cells were analyzed in culture for **DNA** synthesis as measured by 24-hour 5-bromo-2-deoxyuridine (BrdU) uptake. Reverse-transcription **polymerase** chain **reaction** (RT-**PCR**) with **gene**-specific oligonucleotide primers was performed to characterize the PACAP/VIP receptor subtype(s). RESULTS: PACAP/VIP neuropeptide-stimulated BrdU uptake was significantly greater (3.4-3.8-fold greater than control) than that at the maximal dose of gastrin (2.2-fold greater than control). PACAP-stimulated ECL cell

proliferation (EC50, approximately 3 x 10(-)14 mol/L) was approximately 100-fold more potent than VIP (EC50, approximately 3x 10(-)12 mol/L). The stimulated BrdU uptake by both PACAP and VIP was competitively inhibited by PACAP-receptor antagonist (IC50, 10(-)9 mol/L, 3 x 10(-)9 mol/L, respectively) and VIP-receptor antagonist (IC50, 3 x 10(-)7 mol/L, 5 x 10(-)7 mol/L, respectively). RT-**PCR** identified the presence of the PACAP-specific but not PACAP/VIP receptor subtypes. The PACAP-stimulated BrdU uptake was inhibited (70%-80%) by inhibitors of adenosine 3',5'-cyclic monophosphate, **phosphatidylinositol 3 kinase**, and **protein** tyrosine kinase as well as mitogen-activated **protein** kinase. CONCLUSIONS: PACAP/VIP-related **peptides** are more potent modulators of ECL cell proliferation than gastrin, and their effect is mediated by a PACAP-specific receptor whose activation is transduced by multiple intracellular messenger systems.

L21 ANSWER 4 OF 59 MEDLINE DUPLICATE 2
ACCESSION NUMBER: 1999265490 MEDLINE
DOCUMENT NUMBER: 99265490
TITLE: Mutational analysis of the coding regions of the **genes** encoding **protein** kinase B-alpha and -beta, phosphoinositide-dependent **protein** kinase-1, phosphatase targeting to glycogen, **protein** phosphatase inhibitor-1, and glycogenin: lessons from a search for genetic variability of the insulin-stimulated glycogen synthesis pathway of skeletal muscle in NIDDM patients.
AUTHOR: Hansen L; Fjordvang H; Rasmussen S K; Vestergaard H; Echwald S M; Hansen T; Alessi D; Shenolikar S; Saltiel A R; Barbetti F; Pedersen O
CORPORATE SOURCE: Steno Diabetes Center and Hagedorn Research Institute, Gentofte, Denmark.. larh@hagedorn.dk
SOURCE: DIABETES, (1999 Feb) 48 (2) 403-7.
Journal code: E8X. ISSN: 0012-1797.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199908
ENTRY WEEK: 19990802
AB The finding of a reduced insulin-stimulated glucose uptake and glycogen synthesis in the skeletal muscle of glucose-tolerant first-degree relatives of patients with NIDDM, as well as in cultured fibroblasts and skeletal muscle cells isolated from NIDDM patients, has been interpreted as evidence for a genetic involvement in the disease. The mode of inheritance of the common forms of NIDDM is as yet unclear, but the prevailing hypothesis supports a

polygenic model. In the present study, we tested the hypothesis that the putative inheritable defects of insulin-stimulated muscle glycogen synthesis might be caused by genetic variability in the **genes** encoding **proteins** shown by biochemical evidence to be involved in insulin-stimulated glycogen synthesis in skeletal muscle. In 70 insulin-resistant Danish NIDDM patients, mutational analysis by reverse transcription-**polymerase** chain **reaction**-single strand conformation polymorphism-heteroduplex analysis was performed on genomic **DNA** or skeletal muscle-derived cDNAs encoding glycogenin, **protein** phosphatase inhibitor-1, phophatase targeting to glycogen, **protein** kinase B-alpha and -beta, and the **phosphoinositide**-dependent **protein** kinase-1. Although a number of silent variants were identified in some of the examined **genes**, we found no evidence for the hypothesis that the defective insulin-stimulated glycogen synthesis in skeletal muscle in NIDDM is caused by structural changes in the **genes** encoding the known components of the insulin-sensitive glycogen synthesis pathway of skeletal muscle.

L21 ANSWER 5 OF 59 BIOSIS COPYRIGHT 1999 BIOSIS
ACCESSION NUMBER: 1999:125230 BIOSIS
DOCUMENT NUMBER: PREV199900125230
TITLE: Growth arrest-specific **gene** 6 (Gas6)/adhesion related kinase (Ark) signaling promotes gonadotropin-releasing hormone neuronal survival via extracellular signal-regulated kinase (ERK) and Akt.
AUTHOR(S): Allen, Melissa P.; Zeng, Chan; Schneider, Kristina; Xiong, Xiaoyan; Meintzer, Mary Kay; Bellosta, Paola; Basilico, Claudio; Varnum, Brian; Heidenreich, Kim A.; Wierman, Margaret E. (1)
CORPORATE SOURCE: (1) Endocrinol., Veterans Affairs Med. Cent., 1055 Clermont St., Denver, CO 80220 USA
SOURCE: Molecular Endocrinology, (Feb., 1999) Vol. 13, No. 2, pp. 191-201.
ISSN: 0888-8809.
DOCUMENT TYPE: Article
LANGUAGE: English
AB We identified Ark, the mouse homolog of the receptor tyrosine kinase AxI (Ufo, Tyro7), in a screen for novel factors involved in GnRH neuronal migration by using differential-display **PCR** on cell lines derived at two windows during GnRH neuronal development. Ark is expressed in Gn10 GnRH cells, developed from a tumor in the olfactory area when GnRH neurons are migrating, but not in GT1-7 cells, derived from a tumor in the forebrain when GnRH neurons are postmigratory. Since Ark (AxI) signaling protects from programmed cell death in fibroblasts, we hypothesized that it may play an antiapoptotic role in GnRH neurons. Gn10 (Ark positive) GnRH cells

were more resistant to serum withdrawal-induced apoptosis than GT1-7 (Ark negative) cells, and this effect was augmented with the addition of Gas6, the Ark (AxI) ligand. Gas6/Ark stimulated the extracellular signal-regulated kinase, ERK, and the serine-threonine kinase, Akt, a downstream component of the **phosphoinositide** 3-kinase (PI3-K) pathway. To determine whether ERK or Akt activation is required for the antiapoptotic effects of Gas6/Ark in GnRH neurons, cells were serum starved in the absence or presence of Gas6, with or without inhibitors of ERK and PI-13 K signaling cascades. Gas6 rescued Gn10 cells from apoptosis, and this effect was blocked by coincubation of the cells with the mitogen-activated **protein**/ERK kinase (MEK) inhibitor, PD98059, or wortmannin (but not rapamycin). These data support an important role for Gas6/Ark signaling via the ERK and PI3-K (via Akt) pathways in the protection of GnRH neurons from programmed cell death across neuronal migration.

L21 ANSWER 6 OF 59 BIOTECHDS COPYRIGHT 1999 DERWENT INFORMATION LTD
ACCESSION NUMBER: 1998-09554 BIOTECHDS
TITLE: New **nucleic** acid encoding phosphoinositol-kinase-3-C2-alpha or its fragments; human recombinant enzyme preparation by vector expression in host cell, humanized antibody, **DNA** primer, agonist and antisense **nucleic** acid, used for cancer diagnosis, therapy or **gene** therapy
AUTHOR: Domin J; Waterfield M D
PATENT ASSIGNEE: Ludwig-Inst.Cancer-Res.
LOCATION: London, UK.
PATENT INFO: WO 9832864 30 Jul 1998
APPLICATION INFO: WO 1998-GB244 27 Jan 1998
PRIORITY INFO: GB 1997-1652 28 Jan 1997
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 1998-427960 [36]
AN 1998-09554 BIOTECHDS
AB A new and specified 5,062 bp **DNA** sequence encodes human **phosphoinositol**-kinase-3-C2-alpha (I) with a specified 1,686 amino acid **protein** sequence (or a fragment). Also claimed are: **DNA** that hybridizes with the new sequence; (I) or a variant encoded by the **DNA**; any **protein** that is a class-II kinase insensitive to at least one of wortmannin and LY294002 (2-(4-morpholino)-8-phenyl-4H-1-benzopyran-4-one); vectors containing the **DNA**; cells transformed or transfected with the vectors; antibodies against (I); vectors that express an in-frame fusion of human immunoglobulin constant region and rodent immunoglobulin variable region, from a heavy or light chain with specificity of an antibody; **DNA** primers for **polymerase** chain **reaction** of the **DNA**; a

method for identifying (I) agonists and antagonists; and antisense **nucleic** acid. The antibodies, optionally humanized may be used to identify (I), and antisense sequences, antibodies or dominant negative mutants of (I) may be used in veterinary medicine to block (I) or to treat tumor cells where the phenotype is associated with expression of (I). (52pp)

L21 ANSWER 7 OF 59 EMBASE COPYRIGHT 1999 ELSEVIER SCI. B.V.
ACCESSION NUMBER: 1999134354 EMBASE
TITLE: Signaling mechanisms underlying muscarinic receptor-mediated increase in contraction rate in cultured heart cells.
AUTHOR: Colecraft H.M.; Egamino J.P.; Sharma V.K.; Sheu S.-S.
CORPORATE SOURCE: S.-S. Sheu, Dept. of Pharmacology and Physiology, University of Rochester, School of Medicine and Dentistry, Rochester, NY 14642, United States. sheus@pharmacol.rochester.edu
SOURCE: Journal of Biological Chemistry, (27 Nov 1999) 273/48 (32158-32166).
Refs: 44
ISSN: 0021-9258 CODEN: JBCHA3
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English

AB We have investigated the mechanisms by which stimulation of cardiac muscarinic receptors result in paradoxical stimulatory effects on cardiac function, using cultured neonatal rat ventricular myocytes as a model system. Application of low concentrations of carbachol (CCh) (EC50 = 35 nM) produced an atropine-sensitive decrease in spontaneous contraction rate, while, in cells pretreated with pertussis toxin, higher concentrations of CCh (EC50 = 26 .mu.M) elicited an atropine-sensitive increase in contraction rate. Oxotremorine, an m2 muscarinic acetylcholine receptor (mAChR) agonist, mimicked the negative but not the positive chronotropic response to CCh. Reverse transcription followed by **polymerase** chain **reaction** carried out on mRNA obtained from single cells indicated that ventricular myocytes express mRNA for the m1, m2, and, possibly, m4 mAChRs. The presence of m1 and m2 mAChR **protein** on the surface membranes of the cultured ventricular myocytes was confirmed by immunofluorescence. The CCh-induced positive chronotropic response was significantly inhibited by fluorescein-tagged antisense oligonucleotides directed against the m1, but not the m2 and m4, mAChR subtypes. The response was also inhibited by antisense oligonucleotides against G(q).alpha. **protein**. Finally, inhibition of CCh-induced **phosphoinositide** hydrolysis with 500 .mu.M neomycin or 5 .mu.M U73122 completely abolished the CCh-induced positive

chronotropic response. These results are consistent with the stimulatory effects of mAChR activation on the rate of contractions in cultured ventricular myocytes being mediated through the m1 mAChR coupled through G(q) to phospholipase C-induced **phosphoinositide** hydrolysis.

L21 ANSWER 8 OF 59 JICST-EPlus COPYRIGHT 1999 JST
ACCESSION NUMBER: 980822145 JICST-EPlus
TITLE: A single bout of exercise induces a transient alteration in **gene** expression of the insulin signaling pathway.
AUTHOR: INOUE T
KIM Y-B
NAKAJIMA R; TOKUYAMA K; SUZUKI M
SEKINE T
CORPORATE SOURCE: Kure Kyousai Hospital, Hiroshima, Jpn
Harvard Medical School, Ma
Univ. Tsukuba, Ibaraki, Jpn
National Inst. Health & Nutrition, Tokyo, Jpn
SOURCE: Tairyoku Kagaku (Japanese Journal of Physical Fitness and Sports Medicine), (1998) vol. 47, no. 4, pp. 421-426. Journal Code: Z0388B (Fig. 2, Tbl. 1, Ref. 25)
ISSN: 0039-906X
PUB. COUNTRY: Japan
DOCUMENT TYPE: Journal; Article
LANGUAGE: English
STATUS: New
AB The present study was conducted to examine whether **gene** expression of the insulin signaling pathway is activated in response to a single bout of exercise. Adult male rats underwent a single bout of treadmill exercise for 90min at 22m/min on a 6.DEG.C. incline. The animals were sacrificed immediately after exercise or after a predetermined post-exercise recovery period of up to 48h, and the level of mRNA in the gastrocnemius muscle was measured by the RT-**PCR** method. Immediately after exercise, mRNA levels of insulin signal transduction intermediates in the gastrocnemius muscle were lower in the exercise group than in the controls. Following this tendency reduction, mRNA levels of insulin receptor, IRS-1, **PI3-kinase**, SH-PTP2 and Nck were higher in the exercise group than in the controls. The levels of **PI3-kinase** mRNA was significantly higher in the exercise group than in the controls within 6h after exercise, while levels of mRNA for insulin receptor, IRS-1, SH-PTP2 and Nck were significantly higher in the exercise group than in the controls at 48h after exercise. **Gene** expression of oncogenes was also affected by a single bout of exercise. Levels of c-fos and c-myc mRNA were significantly higher than in the controls immediately after exercise, while the level of c-jun mRNA was lower in the

exercise group at 3h after exercise. Thus, a single bout of exercise affects the **gene** expression of insulin signal transduction pathway intermediates and oncogenes in rat gastrocnemius muscle. The present findings suggest that an exercise-induced adaptive increase of skeletal muscle insulin sensitivity is a cumulative effect of a single bout of exercise on the **gene** expression of insulin signal transduction pathway intermediates. (author abst.)

L21 ANSWER 9 OF 59 MEDLINE
ACCESSION NUMBER: 1998193143 MEDLINE
DOCUMENT NUMBER: 98193143
TITLE: Downregulated expression of the signaling molecules Nck, c-Crk, Grb2/Ash, PI 3-kinase p110 alpha and WRN during fibroblast aging in vitro.
AUTHOR: Matuoka K; Takenawa T
CORPORATE SOURCE: Department of Biosignal Research, Tokyo Metropolitan Institute of Gerontology, Japan..
kmatuoka@mailhost.net
SOURCE: BIOCHIMICA ET BIOPHYSICA ACTA, (1998 Feb 4) 1401 (2) 211-5.
Journal code: A0W. ISSN: 0006-3002.
PUB. COUNTRY: Netherlands
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199806
ENTRY WEEK: 19980603
AB An RT-**PCR** analysis was performed to examine changes in intracellular signal transducing molecules during in-vitro aging of human fibroblasts. Expression of Nck, c-Crk, Grb2/Ash, **phosphoinositide** (PI) 3-kinase p110 alpha and Werner's syndrome **gene** product WRN was noticeably reduced in late passage cells, showing a concurrent downregulation of a set of signaling molecules accompanying aging.

L21 ANSWER 10 OF 59 MEDLINE DUPLICATE 3
ACCESSION NUMBER: 1999031965 MEDLINE
DOCUMENT NUMBER: 99031965
TITLE: Chemosignal transduction in the vomeronasal organ of garter snakes: cloning of a **gene** encoding adenylate cyclase from the vomeronasal organ of garter snakes.
AUTHOR: Liu W; Wang D; Liu J; Chen P; Halpern M
CORPORATE SOURCE: Department of Biochemistry, SUNY Health Science Center at Brooklyn, Brooklyn, New York 11203, USA.
CONTRACT NUMBER: DC 00104 (NIDCD)
SOURCE: ARCHIVES OF BIOCHEMISTRY AND BIOPHYSICS, (1998 Oct 15) 358 (2) 204-10.
Journal code: 6SK. ISSN: 0003-9861.
Searcher : Shears 308-4994

PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199901
ENTRY WEEK: 19990104

AB We previously reported that ES20-receptor binding activates **phosphoinositide** (PI) turnover, resulting in an increase in inositol-1,4,5-trisphosphate, which in turn mobilizes intracellularly stored calcium in the vomeronasal (VN) sensory epithelium of garter snakes. We also found that the activity of adenylate cyclase (AC) in the VN organ is very sensitive to Ca2+ but insensitive to calmodulin regulation. A 250-bp fragment of adenylate cyclase type VI (AC-VI) was obtained from brain cDNA of garter snake by RT-**PCR** with degenerate primers. The 250-bp fragments were amplified, cloned, and sequenced. Both Northern blot and RNase protection assays revealed that the vomeronasal organ (VNO) and brain contained more abundance of AC type VI than the main olfactory epithelium. A 3.8-kb cDNA was then cloned from the vomeronasal cDNA library of garter snakes and sequenced. The 5' cDNA was obtained by means of 5' RACE **PCR** and sequenced. We have successfully cloned a 5200-nucleotide cDNA from VNO of garter snakes containing an open reading frame++ encoding 1150 amino acids of AC-VI **protein**. The vomeronasal AC is termed AC(VN) . AC(VN) shows a high degree of homology with type VI AC of rat, mouse, or human. In situ hybridization with digoxigenin-labeled cRNA demonstrated that AC(VN) mRNA was abundant in the sensory epithelium but not in the nonsensory epithelium of the mushroom body of the vomeronasal organ of garter snakes. Copyright 1998 Academic Press.

L21 ANSWER 11 OF 59 JICST-EPlus COPYRIGHT 1999 JST
ACCESSION NUMBER: 990118726 JICST-EPlus
TITLE: Thromboxane A2 receptor-mediated signal transduction in glial cells.
AUTHOR: NAKAHATA NORIMICHI; HONMA SHIGEYOSHI; KOBAYASHI HIROSHI; OIZUMI YASUSHI
CORPORATE SOURCE: Tohoku Univ., Fac. of Pharm.
SOURCE: Nippon Yakurigaku Zasshi (Folia Pharmacologica Japonica), (1998) vol. 112, pp. 113P-117P. Journal Code: G0740A (Fig. 4, Ref. 19)
CODEN: NYKZAU; ISSN: 0015-5691
PUB. COUNTRY: Japan
DOCUMENT TYPE: Journal; Article
LANGUAGE: Japanese
STATUS: New

AB Thromboxane A2(TXA2) receptor subtypes and their signal transduction were examined in 1321N1 human astrocytoma cells. Placental and endothelial types of TXA2 receptor mRNA were found in astrocytoma cells by RT-**PCR** procedure. Using immunoaffinity column

conjugated with anti-TXA2 receptor antibody, two TXA2 receptors(58 and 55 kDa)were partially purified. The partially purified TXA2 receptor fraction contained Gq/11 and G12. The incubation of the cells with dibutyryl cyclic AMP(dbcAMP) caused a differentiation of human astrocytoma cells. DbcAMP treatment resulted in the reduction of Ca2+ elevation and **phosphoinositide** hydrolysis induced by TXA2 receptor agonist. Whereas the responsiveness of Ca2+ signaling was weakened by dbcAMP treatment, phosphorylation of mitogen-activated **protein** kinase was increased in dbcAMP-treated cells. These results suggest that human astrocytoma cells express placental and endothelial TXA2 receptors. The TXA2 receptors were coupled with Gq/11 and G12. DbcAMP treatment discriminates TXA2 receptor-mediated MAPK activation from the Ca2+ signaling pathway. (author abst.)

L21 ANSWER 12 OF 59 MEDLINE DUPLICATE 4
ACCESSION NUMBER: 1998081734 MEDLINE
DOCUMENT NUMBER: 98081734
TITLE: Depot-specific differences in adipose tissue **gene** expression in lean and obese subjects.
AUTHOR: Lefebvre A M; Laville M; Vega N; Riou J P; van Gaal L; Auwerx J; Vidal H
CORPORATE SOURCE: INSERM U.325, Departement d'Atherosclerose, Institut Pasteur, Lille, France.
SOURCE: DIABETES, (1998 Jan) 47 (1) 98-103.
Journal code: E8X. ISSN: 0012-1797.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199803
ENTRY WEEK: 19980304
AB Intra-abdominal and subcutaneous adipose tissue display important metabolic differences that underlie the association of visceral, but not subcutaneous, fat with obesity-related cardiovascular and metabolic problems. Because the molecular mechanisms contributing to these differences are not yet defined, we compared by reverse transcription-**polymerase** chain **reaction** the expression of 15 mRNAs that encode **proteins** of known importance in adipocyte function in paired omental and subcutaneous abdominal biopsies. No difference in mRNA expression between omental and subcutaneous adipose tissue was observed for hormone sensitive lipase, lipoprotein lipase, 6-phosphofructo-1-kinase, insulin receptor substrate 1, p85alpha regulatory subunit of **phosphatidylinositol-3-kinase**, and Rad.
Total amount of insulin receptor expression was significantly higher in omental adipose tissue. Most of this increase was accounted for by expression of the differentially spliced insulin receptor lacking exon 11, which is considered to transmit the insulin signal less

efficiently than the insulin receptor with exon 11. Perhaps consistent with a less efficient insulin signaling, a twofold reduction in GLUT4, glycogen synthase, and leptin mRNA expression was observed in omental adipose tissue. Finally peroxisome proliferator activated receptor-gamma (PPAR-gamma) mRNA levels were significantly lower in visceral adipose tissue in subjects with a BMI <30 kg/m2, but not in obese subjects, indicating that relative PPAR-gamma expression is increased in omental fat in obesity. This suggests that altered expression of PPAR-gamma might play a role in adipose tissue distribution and expansion.

L21 ANSWER 13 OF 59 EMBASE COPYRIGHT 1999 ELSEVIER SCI. B.V.
ACCESSION NUMBER: 1998067479 EMBASE
TITLE: Mechanical stretch/relaxation stimulates a cellular renin-angiotensin system in cultured rat mesangial cells.
AUTHOR: Becker B.N.; Yasuda T.; Kondo S.; Vaikunth S.; Homma T.; Harris R.C.
CORPORATE SOURCE: Dr. R.C. Harris, Division of Nephrology, Vanderbilt Univ. School of Medicine, S-3223 Medical Center North, Nashville, TN 37232-2372, United States
SOURCE: Experimental Nephrology, (1998) 6/1 (57-66).
Refs: 49
ISSN: 1018-7782 CODEN: EXNEEG
COUNTRY: Switzerland
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 003 Endocrinology
028 Urology and Nephrology
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Angiotensin II (Ang II) may play a significant role mediating intraglomerular hypertension and glomerular sclerosis. Therefore, we investigated whether a model of pressure-induced stress, mechanical stretch/relaxation, affected the renin-angiotensin system (RAS) in cultured rat mesangial cells. Type 1 Ang II receptor (AT1R) expression was assessed by 125I-Ang II binding and quantitative reverse-transcription **polymerase** chain **reaction**. Stretch/relaxation increased steady-state AT1R mRNA levels as well as specific [125I]Ang II binding. Increased AT.apprx.R expression was associated with altered AT1R signaling. Ang II (100 nM) increased total **phosphoinositide** hydrolysis in control cells (186 .+-. 25%, n = 6; p < 0.025 vs. no treatment). However, stretch/relaxation for 48 h further augmented AT 1R-mediated PI hydrolysis (293 .+-. 38%, n = 6; p < 0.025 vs. Ang II treatment alone). We examined other RAS components in mesangial cells subjected to stretch/relaxation. Angiotensinogen, determined by radioimmunoassay of Ang I generation in conditioned media, increased with stretch/relaxation, and reverse-transcription **polymerase** chain **reaction** demonstrated increased

angiotensinogen **gene** expression in stretch/relaxation-treated cells. However, renin activity and angiotensin-converting-enzyme-like activity were unaffected by stretch/relaxation. Thus, mesangial cells maintain a local RAS similar to those described in other tissues, and AT1R expression and angiotensinogen production in this cellular RAS are increased by stretch/relaxation. It is likely that mesangial cells in vivo, exposed to variations in intraglomerular pressure, may regulate their responses via a local RAS.

L21 ANSWER 14 OF 59 MEDLINE DUPLICATE 5
ACCESSION NUMBER: 1998129081 MEDLINE
DOCUMENT NUMBER: 98129081
TITLE: Genetic characterization of a phospholipase C **gene** from Candida albicans: presence of homologous sequences in Candida species other than Candida albicans.
AUTHOR: Bennett D E; McCreary C E; Coleman D C
CORPORATE SOURCE: University of Dublin, School of Dental Science, Department of Oral Medicine and Pathology, Trinity College, Dublin, Republic of Ireland.
SOURCE: MICROBIOLOGY, (1998 Jan) 144 (Pt 1) 55-72. Journal code: BXW. ISSN: 1350-0872.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-Y13975; GENBANK-Y14062
ENTRY MONTH: 199806
ENTRY WEEK: 19980602

AB Phospholipase C (PLC) enzymes are essential in regulating several important cellular functions in eukaryotes, including yeasts. In this study, **PCR** was used to identify a **gene** encoding PLC activity in Candida albicans, using oligonucleotide primers complementary to sequences encoding highly conserved amino acid regions within the X domains of previously characterized eukaryotic phospholipase C **genes**. The nucleotide sequence of the C. albicans **gene**, CAPLC1 (2997 bp), was determined from a recombinant clone containing C. albicans 132A genomic **DNA**; it encoded a **polypeptide** of 1099 amino acids with a predicted molecular mass of 124.6 kDa. The deduced amino acid sequence of this **polypeptide** (CAPLC1) exhibited many of the features common to previously characterized PLCs, including specific X and Y catalytic domains. The CAPLC1 **protein** also exhibited several unique features, including a novel stretch of 18-19 amino acid residues within the X domain and an unusually long N-terminus which did not contain a recognizable EF-hand Ca(2+)-binding domain. An overall amino acid homology of more than 27% with PLCs previously characterized from Saccharomyces cerevisiae

and Schizosaccharomyces pombe suggested that the CAPLC1 **protein** is a delta-form of **phosphoinositide** -specific PLC (PI-PLC). PLC activity was detected in cell-free extracts of both yeast and hyphal forms of C. albicans 132A following 7 h and 24 h growth using the PLC-specific substrate p-nitrophenylphosphorylcholine (p-NPPC). In addition, CAPLC1 mRNA was detected by reverse transcriptase **PCR** in both yeast and hyphal forms of C. albicans 132A at the same time intervals. Expression of CAPLC1 activity was also detected in extracts of Escherichia coli DH5 alpha harbouring plasmids which contained portions of the CAPLC1 **gene** lacking sequences encoding part of the N-terminus. Southern hybridization and **PCR** analyses revealed that all C. albicans and Candida dubliniensis isolates examined possessed sequences homologous to CAPLC1. Sequences related to CAPLC1 were detected in some but not all isolates of Candida tropicalis, Candida glabrata and Candida parapsilosis tested, but not in the isolates of Candida krusei, Candida kefyr, Candida guillermondii and Candida lusitaniae examined. This paper reports the first description of the cloning and sequencing of a PLC **gene** from a pathogenic yeast species.

L21 ANSWER 15 OF 59 BIOTECHDS COPYRIGHT 1999 DERWENT INFORMATION LTD
ACCESSION NUMBER: 1997-12151 BIOTECHDS
TITLE: New isolated phosphatidyl inositol-3 **polypeptide**; 1-phosphatidylinositol-3-kinase preparation for use in proliferative disorder therapy
AUTHOR: Williams L T; Molz L; Chen Y W
PATENT ASSIGNEE: Univ.California
LOCATION: Oakland, CA, USA.
PATENT INFO: WO 9731650 4 Sep 1997
APPLICATION INFO: WO 1997-US2193 12 Feb 1997
PRIORITY INFO: US 1996-609049 29 Feb 1996
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 1997-448442 [41]
AN 1997-12151 BIOTECHDS
AB A new 1-**phosphatidylinositol**-3-**kinase** (**PI3-kinase**, EC-2.7.1.37), derived from Drosophila sp. or a mouse, is encoded by a **nucleic** acid (**DNA** sequence and **protein** sequence specified) which can be contained on a vector and used to transform a bacterial, mammal, plant, fungal or insect host cell, preferably a COS-7 cell. An antibody specifically reactive with a **PI3** -**kinase** can inhibit its action. The **PI3**-**kinase** can be used to treat a patient with symptoms of a disorder caused by dysregulation of a growth factor activation signalling cascade. In particular, products and methods disclosed

can be used in diagnosis, drug screening and treatment of proliferative disorders including atherosclerosis, inflammatory joint diseases, psoriasis, restenosis following angioplasty, and cancer. Also claimed is a **DNA** probe with a specified sequence. The **genes** are obtained by **polymerase** chain **reaction** amplification of Drosophila and mouse cDNA libraries with **DNA** primers. (77pp)

L21 ANSWER 16 OF 59 BIOSIS COPYRIGHT 1999 BIOSIS
ACCESSION NUMBER: 1997:313982 BIOSIS
DOCUMENT NUMBER: PREV199799604470
TITLE: A truncated form of RGS3 negatively regulates G **protein**-coupled receptor stimulation of adenylyl cyclase and phosphoinositide phospholipase C.
AUTHOR(S): Chatterjee, Tapan K.; Eapen, Alex K.; Fisher, Rory A. (1)
CORPORATE SOURCE: (1) Dep. Pharmacol., Univ. Iowa Coll. Med., Iowa City, IA 52242 USA
SOURCE: Journal of Biological Chemistry, (1997) Vol. 272, No. 24, pp. 15481-15487.
ISSN: 0021-9258.
DOCUMENT TYPE: Article
LANGUAGE: English
AB Identification of a new family of **proteins** (RGS **proteins**) that function as negative regulators of G **protein** signaling has sparked new understanding of desensitization of this signaling process. Recent studies with several mammalian RGS **proteins** has delineated their ability to interact with and function as GTPase-activating **proteins** specifically for G **proteins** in the G-i family. Here, we investigated the functional activity of RGS3 and a truncated form of RGS3 on G **protein**-coupled receptor-mediated activation of adenylyl cyclase, **phosphoinositide** phospholipase C, and mitogen-activated **protein** kinase in intact cells. **Polymerase** chain **reaction** and 5'-rapid amplification of cDNA ends analyses revealed the tissue-specific expression of a short form of the RGS3 transcript that encodes the approximate carboxyl-terminal half of RGS3. This truncated form of RGS3 (RGS3T) was shown recently to function as a negative regulator of pheromone signaling in yeast (Druey, IC M., Blumer, K. J., Hang, V. R., and Kehrl, J. H. (1996) Nature 379, 742-746). Baby hamster kidney cells transiently transfected with RGS3T cDNA exhibited a pronounced impairment in platelet-activating factor receptor-stimulated inositol phosphate production, a pertussis toxin-insensitive response. Similarly, calcitonin **gene**-related **peptide** receptor-stimulated increases in intracellular cAMP and pituitary adenylate-cyclase activating **polypeptide**

receptor-stimulated increases in both cAMP and inositol phosphates were reduced significantly in RGS3T transfectants compared with vector-transfected control cells. In contrast, baby hamster kidney cells transfected with the full-length RGS3 cDNA showed no impairment in cAMP and inositol phosphate production mediated by these G **protein**-coupled receptors. However, lysophosphatidic acid receptor-stimulated phosphorylation of endogenous ERK1 and ERK2 was impaired markedly in both RGS3 and RGS3T transfectants, demonstrating the functional ability of both RGS forms to modulate G-i-mediated signaling. These results provide the first evidence for regulatory effects of an RGS **protein** on G-s- and G-q-mediated signaling in intact cells and document that the carboxyl-terminal region of RGS3 comprises the structural domain for this activity.

L21 ANSWER 17 OF 59 MEDLINE DUPLICATE 6
ACCESSION NUMBER: 97160567 MEDLINE
DOCUMENT NUMBER: 97160567
TITLE: Human GRB-IRbeta/GRB10. Splice variants of an insulin and growth factor receptor-binding **protein** with PH and SH2 domains.
AUTHOR: Frantz J D; Giorgetti-Peraldi S; Ottinger E A; Shoelson S E
CORPORATE SOURCE: Joslin Diabetes Center & Department of Medicine, Harvard Medical School, Boston, Massachusetts 02215, USA.
CONTRACT NUMBER: DK43123 (NIDDK)
DK45943 (NIDDK)
DK36836 (NIDDK)
+
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (1997 Jan 31) 272 (5) 2659-67.
Journal code: HIV. ISSN: 0021-9258.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199705
AB cDNA clones encoding human (h) Grb7 and a previously unknown **protein** with high homology to hGrb-IR and mGrb10 (where m indicates mouse) were found by screening expressed sequence tag data bases. hGrb7 mRNA expression is greatest in pancreas and restricted to a few other tissues. The second **protein** termed hGrb-IRbeta/Grb10 contains an intact PH domain and lacks the 80-residue mGrb10 insertion. Expression is greatest in pancreas and muscle but occurs in nearly all tissues. hGrb-IRbeta/Grb10 and hGrb-IR likely arise as alternative mRNA splicing products of a common **gene**. Reverse transcriptase-coupled **polymerase** chain **reaction** shows both mRNAs in

muscle. In cells, Grb-IRbeta/Grb10 **protein** translocates from cytosol to membrane upon insulin stimulation, most likely due to direct interactions with the insulin receptor. These interactions are mediated by the SH2 domain and additional regions of the **protein**. Studies with mutated receptors and synthetic phosphopeptides show that the hGrb-IRbeta/Grb10 SH2 domain binds at least two sites in the insulin receptor: the kinase activation loop > the juxtamembrane site. hGrb-IRbeta/Grb10 also binds a 135-kDa phosphoprotein in unstimulated 3T3-L1 adipocytes; binding is reduced upon insulin stimulation. In addition, the c-Abl SH3 domain binds Grb-IR/Grb10, whereas Fyn, **phosphatidylinositol 3 -kinase** p85, and Grb2 SH3 domains do not. The site of c-Abl SH3 domain interaction is highly conserved within the Grb-IR/Grb10/Grb7/Grb14 family. hGrb-IRbeta/Grb10 also binds platelet-derived growth factor and epidermal growth factor receptors, suggesting a broader role in the signaling pathways of numerous receptors. We conclude that hGrb-IRbeta/Grb10 is a widely expressed, PH and SH2 domain-containing, SH3 domain-binding **protein** that functions downstream from activated insulin and growth factor receptors.

L21 ANSWER 18 OF 59 SCISEARCH COPYRIGHT 1999 ISI (R)
ACCESSION NUMBER: 97:248627 SCISEARCH
THE GENUINE ARTICLE: WN858
TITLE: Pituitary adenylate cyclase-activating **polypeptide** triggers dual transduction signaling in CATH.a cells and transcriptionally activates tyrosine hydroxylase and c-fos expression
AUTHOR: Muller A; Monnier D; Rene F; Larmet Y; Koch B; Loeffler J P (Reprint)
CORPORATE SOURCE: URA CNRS 1446, IPCB, LAB NEUROPHYSIOL & NEUROBIOL SYST ENDOCRINES, 21 RUE RENE DESCARTES, F-67084 PARIS, FRANCE (Reprint); URA CNRS 1446, IPCB, LAB NEUROPHYSIOL & NEUROBIOL SYST ENDOCRINES, F-67084 PARIS, FRANCE; HOSPICES CIVILS STRASBOURG, CLIN DOULEUR, STRASBOURG, FRANCE
COUNTRY OF AUTHOR: FRANCE
SOURCE: JOURNAL OF NEUROCHEMISTRY, (APR 1997) Vol. 68, No. 4, pp. 1696-1704.
Publisher: LIPPINCOTT-RAVEN PUBL, 227 EAST WASHINGTON SQ, PHILADELPHIA, PA 19106.
ISSN: 0022-3042.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE
LANGUAGE: English
REFERENCE COUNT: 38
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB We used a catecholaminergic neuron-like cell line (CATH.a cells) as a model system to investigate the likelihood that pituitary

adenylate cyclase-activating **polypeptide** (PACAP) may participate in the regulation of specific **gene** expression in catecholaminergic neurons. Analysis by reverse transcriptase-**PCR** amplification revealed the presence in these cells of type I PACAP receptors, with a short isoform, together with a heavier so-called Hop splice variant. PACAP38 and PACAP27 enhanced, in a dose-dependent manner, both cyclic AMP formation and **phosphoinositide** breakdown, with EC(50), values of, respectively, 0.6 x 10(-10) and 2 x 10(-9) M. These **peptides**, in addition, also elevated [Ca2+](i) by mobilizing intracellular calcium pools. Vasoactive intestinal **peptide** (VIP) was similar to 1,000-fold less potent in stimulating cyclic AMP (with EC(50) = 2 x 10(-7) M) and failed to change the turnover of **phosphoinositides** and to alter [Ca2+](i). Both forms of PACAP, as well as forskolin, stimulated transcriptional induction of tyrosine hydroxylase (TH) and c-fos promoters fused to a chloramphenicol acetyltransferase (CAT) reporter **gene** in transiently transfected cells ($p < 0.01$ vs. controls). Induction of CAT activity linked to both TH and c-fos promoters was obliterated upon coexpression of a dominant inhibitory mutant (Mt-RAB) of cyclic AMP-dependent **protein** kinase. We conclude that CATH.a cells do express functional PACAP type I receptors, the activation of which impinges on TH and c-fos transcription according to a process that is primarily dependent on the cyclic AMP-PKA pathway.

L21 ANSWER 19 OF 59 MEDLINE
ACCESSION NUMBER: 97306430 MEDLINE
DOCUMENT NUMBER: 97306430
TITLE: Identification of putative phosphoinositide-specific phospholipase C **genes** in filamentous fungi.
AUTHOR: Jung O J; Lee E J; Kim J W; Chung Y R; Lee C W
CORPORATE SOURCE: Department of Microbiology, College of Natural Sciences, Gyeongsang National University, Chinju, Korea.
SOURCE: MOLECULES AND CELLS, (1997 Apr 30) 7 (2) 192-9.
Journal code: CRQ. ISSN: 1016-8478.
PUB. COUNTRY: KOREA
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-U65685; GENBANK-U65684; GENBANK-U65686; GENBANK-U65687; GENBANK-U65688
ENTRY MONTH: 199710
AB Five putative **phosphoinositide**-specific phospholipases C (PLC) **genes** were identified in three species of filamentous fungi. Using **polymerase** chain **reaction** with degenerate oligonucleotide primers, **gene** fragments encoding amino acid sequences homologous to PLCs of mammals and other organisms were amplified: one sequence from Botryotinia

fuckeliana, one from Aspergillus nidulans, and three from Neurospora crassa. The molecular cloning and sequencing of a putative PLC **gene** (BCPLC1) from B. fuckeliana showed that it encoded a **polypeptide** containing X and Y domains, the two conserved regions found in all known PLCs. The hypothetical **gene** product of BCPLC1 was of delta type in its primary structural organization. The identification of three PLC **genes** in N. crassa shows that multiple PLC isozymes also occur in microbial eukaryotes.

L21 ANSWER 20 OF 59 EMBASE COPYRIGHT 1999 ELSEVIER SCI. B.V.DUPLICATE 7

ACCESSION NUMBER: 97092472 EMBASE
DOCUMENT NUMBER: 1997092472
TITLE: Tumor necrosis factor-.alpha. induces interleukin-6 mRNA and **protein** in human granulosa luteinizing cells via **protein** tyrosine kinase without involving ceramide.
AUTHOR: Machelon V.; Nome F.; Durand-Gasselin I.; Emilie D.
CORPORATE SOURCE: V. Machelon, INSERM, U-355, Maturation Gametique et Fecondation, 32 rue des Carnets, 92140 Clamart, France
SOURCE: Molecular and Cellular Endocrinology, (1997) 126/2 (173-184).
Refs: 50
ISSN: 0303-7207 CODEN: MCEND6
PUBLISHER IDENT.: S 0303-7207(96)03985-8
COUNTRY: Ireland
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 003 Endocrinology
026 Immunology, Serology and Transplantation
029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English

AB This study examines how interleukin-6 (IL-6) expression by human luteinizing granulosa cells is regulated. IL-6 was assayed in culture supernatants, mRNA in cells by in situ hybridization and by a competitive reverse-transcriptase **polymerase** chain **reaction** (RT-**PCR**). TNF.alpha. (100 pg-1 ng/ml) induced IL-6 mRNA and **protein**. Phorbol 12-myristate 13-acetate (PMA) (50 nM) mimicked this effect. DibutyrylcAMP (1 mM) and 10 .mu.M forskolin, C2-, C6- and C8-ceramide (15 .mu.M), all had no effect. The inhibitor of **protein** tyrosine kinase (PTK), genistein (100 .mu.g/ml) reduced tumor necrosis factor (TNF) effects. The inhibitors of **protein** kinase C (PKC) (staurosporine, 10 nM), of phospholipase C (U73122, 2 .mu.M), of phospholipase A2 (PLA2), (indomethacin 30 .mu.M, mepacrin 50 .mu.M, nordihydroguaiaretic acid 10 .mu.M, ONO-RS-082 3,5 .mu.M), none prevented it. Hence, IL-6 is induced by TNF.alpha. via activation of

PTK. **Protein** kinase A, **phosphoinositide** and conventional PKC, sphingomyelin and PLA2 pathways are not implicated.

L21 ANSWER 21 OF 59 MEDLINE
ACCESSION NUMBER: 1998087431 MEDLINE
DOCUMENT NUMBER: 98087431
TITLE: The Arabidopsis thaliana genome has multiple divergent forms of phosphoinositol-specific phospholipase C1.
AUTHOR: Hartweck L M; Llewellyn D J; Dennis E S
CORPORATE SOURCE: Commonwealth Scientific and Industrial Research Organisation, Plant Industry, Canberra, ACT, Australia.
SOURCE: GENE, (1997 Nov 20) 202 (1-2) 151-6.
Journal code: FOP. ISSN: 0378-1119.
PUB. COUNTRY: Netherlands
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-U76423
ENTRY MONTH: 199803
ENTRY WEEK: 19980304

AB Highly degenerate primers to conserved regions of the eukaryotic **phosphoinositol**-specific phospholipase C (PLC) were used to amplify fragments of plant PLCs from Arabidopsis thaliana genomic **DNA**. Eight completely different fragment sequences that showed high homology to PLCs of both animals and plants were isolated. The variation between these putative PLCs was high and suggests that, like animals, plants have multiple isoforms of PLC. Using one of the **PCR** clones, we isolated a corresponding full-length Arabidopsis PLC **gene** (ATHATPLC1G), and sequence analysis indicated that it was most like a delta-type PLC. This **gene** is 2.5 kb and contains seven introns, all but one of which has intron/exon border sequences that conform to the Arabidopsis consensus. The structural complexity of the **gene** is relatively simple compared to mammalian beta-type PLCs that can be 15 kb long with up to 30 introns. The plant **gene** is a single copy and was mapped to four Arabidopsis YACs, one located on chromosome 2. The promoter region contained two TATA-like elements at -43 and -185 and other putative regulatory elements that suggest that this PLC is hormonally regulated. This is the first plant PLC **gene** and the first delta type-PLC **gene** from a higher organism to be sequenced.

L21 ANSWER 22 OF 59 MEDLINE DUPLICATE 8
ACCESSION NUMBER: 97190972 MEDLINE
DOCUMENT NUMBER: 97190972
TITLE: Endothelin ETA receptor regulates signaling and ANF

gene expression via multiple G protein-linked pathways.

AUTHOR: Hilal-Dandan R; Ramirez M T; Villegas S; Gonzalez A; Endo-Mochizuki Y; Brown J H; Brunton L L

CORPORATE SOURCE: Department of pharmacology, University of California, San Diego, La Jolla 92093, USA.

CONTRACT NUMBER: HL-41307 (NHLBI)
HL-2813 (NHLBI)
HL-46345 (NHLBI)
+

SOURCE: AMERICAN JOURNAL OF PHYSIOLOGY, (1997 Jan) 272 (1 Pt 2) H130-7.
Journal code: 3U8. ISSN: 0002-9513.

PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 199706

AB We have characterized the interaction of endothelin (ET) with cultured neonatal rat ventricular myocytes. Binding studies indicate a single population of ETA receptors [53,000 sites/cell, apparent dissociation constant (Kd) for ET-1 approximately 0.07 nM]. Analysis of mRNA levels for ET receptors using 35 cycles of reverse transcriptase-polymerase chain reaction demonstrates the presence of only ETA-receptor message. Studies with ET-1 and a variety of congeners and antagonists indicate that ETA receptors couple to both the stimulation of phosphoinositide turnover and the inhibition of adenylyl cyclase. In myocytes transfected with an atrial natriuretic factor (ANF) promoter linked to a luciferase reporter gene, ET-1 stimulates luciferase expression through an ETA receptor. These data indicate that the ETA receptor is the exclusive receptor on neonatal ventricular myocytes and that this receptor couples to both phosphoinositide hydrolysis and adenylyl cyclase. ET-1 also induces a threefold increase in mitogen-activated protein kinase (MAPK) activity, an effect that is not sensitive to pertussis toxin (PTx). By contrast, ET-stimulated ANF-luciferase expression is partially inhibited by treatment of cells with PTx, suggesting that both PTx-sensitive (Gi) and PTx-insensitive (Gq) pathways mediate the effects of ET-1 on ANF gene expression in neonatal myocytes and that hormonal regulation of ANF expression may utilize pathways in addition to the activation of MAPK.

L21 ANSWER 23 OF 59 MEDLINE

ACCESSION NUMBER: 97339450 MEDLINE

DOCUMENT NUMBER: 97339450

TITLE: Identification of four novel human phosphoinositide 3-kinases defines a multi-isoform subfamily.

AUTHOR: Ho L K; Liu D; Rozycka M; Brown R A; Fry M J

CORPORATE SOURCE: Section of Cell Biology and Experimental Pathology, Institute of Cancer Research, Haddow Laboratories, Sutton, Surrey, United Kingdom.
SOURCE: BIOCHEMICAL AND BIOPHYSICAL RESEARCH COMMUNICATIONS, (1997 Jun 9) 235 (1) 130-7.
Journal code: 9Y8. ISSN: 0006-291X.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199709

AB **Phosphoinositide** (PI) 3-kinases have critical roles in diverse cellular signalling processes and in **protein** trafficking. This suggests that like other intracellular signalling molecules, e.g., phospholipase C and **protein** kinase C, there might be a large family of PI 3-kinase isoforms with the individual members having discrete signalling roles. Reverse transcription-**polymerase** chain **reaction** methods, using degenerate oligonucleotide primers against the lipid kinase consensus region, revealed eight sequences from human cDNA containing a high degree of identity to the family of PI 3-kinases. The sequences obtained included the previously described p110 alpha, p110 beta, and p110 gamma isoforms and HsVps34. Additionally, we have identified four novel sequences which are related to PI 3-kinases. Three of the novel sequences would appear to form a distinct sub-family of PI 3-kinases. We report the expression of these novel PI 3-kinases in human tissues and in cells derived from normal breast.

L21 ANSWER 24 OF 59 EMBASE COPYRIGHT 1999 ELSEVIER SCI. B.V.
ACCESSION NUMBER: 97046333 EMBASE
DOCUMENT NUMBER: 1997046333
TITLE: Endothelin ET(A) receptor regulates signaling and ANF **gene** expression via multiple G **protein**-linked pathways.
AUTHOR: Hilal-Dandan R.; Ramirez M.T.; Villegas S.; Gonzalez A.; Endo-Mochizuki Y.; Brown J.H.; Brunton L.L.
CORPORATE SOURCE: L.L. Brunton, Dept. of Pharmacology 0636, UCSD School of Medicine, San Diego, CA 92093-0636, United States
SOURCE: American Journal of Physiology - Heart and Circulatory Physiology, (1997) 272/1 41-1 (H130-H137).
Refs: 35
ISSN: 0363-6135 CODEN: AJPPDI
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 002 Physiology
029 Clinical Biochemistry
LANGUAGE: English

Searcher : Shears 308-4994

SUMMARY LANGUAGE: English

AB We have characterized the interaction of endothelin (ET) with cultured neonatal rat ventricular myocytes. Binding studies indicate a single population of ET(A) receptors [53,000 sites/cell, apparent dissociation constant (K(d)) for ET-1 = 0.07 nM]. Analysis of mRNA levels for ET receptors using 35 cycles of reverse transcriptase-**polymerase** chain **reaction** demonstrates the presence of only ET(A)-receptor message. Studies with ET-1 and a variety of congeners and antagonists indicate that ET(A) receptors couple to both the stimulation of **phosphoinositide** turnover and the inhibition of adenylyl cyclase. In myocytes transfected with an atrial natriuretic factor (ANF) promoter linked to a luciferase reporter gone, ET-1 stimulates luciferase expression through an ET(A) receptor. These data indicate that the ETA receptor is the exclusive receptor on neonatal ventricular myocytes and that this receptor couples to both **phosphoinositide** hydrolysis and adenylyl cyclase. ET-1 also induces a threefold increase in mitogen-activated **protein** kinase (MAPK) activity, an effect that is not sensitive to pertussis toxin (PTx). By contrast, ET-stimulated ANF-luciferase expression is partially inhibited by treatment of cells with PTx, suggesting that both PTx-sensitive (G(i)) and PTx-insensitive (G(q)) pathways mediate the effects of ET-1 on ANF **gene** expression in neonatal myocytes and that hormonal regulation of ANF express on may utilize pathways in addition to the activation of MAPK.

L21 ANSWER 25 OF 59 LIFESCI COPYRIGHT 1999 CSA

ACCESSION NUMBER: 97:41519 LIFESCI

TITLE: Molecular cloning of a novel variant of the pituitary adenylate cyclase-activating **polypeptide** (PACAP) receptor that stimulates calcium influx by activation L-type calcium channels

AUTHOR: Chatterjee, T.K.; Sharma, R.V.; Fisher, R.A.*

CORPORATE SOURCE: Dep. Pharmacol., Univ. Iowa Coll. Med., Iowa City, IA 52242, USA

SOURCE: J. BIOL. CHEM., (1996) vol. 271, no. 50, pp. 32226-32232.
ISSN: 0021-9258.

DOCUMENT TYPE: Journal

FILE SEGMENT: N; G; T; N3

LANGUAGE: English

SUMMARY LANGUAGE: English

AB Pituitary adenylate cyclase-activating **polypeptide** (PACAP) is a novel neuropeptide that produces its biological effects by interacting with G **protein**-coupled receptors. Molecular cloning of the PACAP receptor revealed the existence of five splice variant receptor forms differing in the third intracellular loop region, with four variants activating both adenylyl cyclase and **phosphoinositide** phospholipase C and one variant activating

only adenylyl cyclase. Here, we report cloning of a novel PACAP receptor variant, designated PACAPR TM4 (transmembrane domain IV), that differs from the previously cloned short form of the PACAP receptor (PACAPR) primarily by discrete sequences located in transmembrane domains II and IV. Reverse transcriptase-**polymerase** chain **reaction** and primer extension analyses demonstrated tissue-specific differential expression of mRNAs encoding PACAPR TM4 and splice variant forms of the PACAP receptor. PACAPR TM4 and PACAPR possess identical intracellular domains, implicated as primary determinants of G **protein** recognition by rhodopsin-like receptors. However, unlike the PACAPR, PACAPR TM4 does not activate either adenylyl cyclase or **phosphoinositide** phospholipase C in response to PACAP in either transient or stable expression systems. However, PACAP stimulates increases in [Ca super(2+)] sub(i) in cells expressing PACAPR TM4 by activating L-type Ca super(2+) channels, a response not elicited by stimulation with vasoactive intestinal **polypeptide**. The signaling phenotype of PACAPR TM4 is characteristic of the PACAP receptor involved in regulation of insulin secretion from pancreatic beta islets, a tissue expressing transcripts for PACAPR TM4 but not for PACAPR or its longer splice variant forms. These findings are consistent with a role of PACAPR TM4 in the physiological control of insulin release by PACAP in beta -islet cells. The finding that PACAPR TM4 has a unique signaling phenotype, although it possesses intracellular domains identical to those of the PACAPR, suggests that receptor-G **protein** recognition by rhodopsin-like receptors can be determined by sequences other than those located in intracellular receptor domains.

L21 ANSWER 26 OF 59 LIFESCI COPYRIGHT 1999 CSA
ACCESSION NUMBER: 97:25935 LIFESCI
TITLE: A novel form of the G **protein** beta subunit G beta sub(5) is specifically expressed in the vertebrate retina
AUTHOR: Watson, A.J.; Aragay, A.M.; Slepak, V.Z.; Simon, M.I.*
CORPORATE SOURCE: Div. Biol., 147-75, California Inst. Technol., Pasadena, CA 91125, USA
SOURCE: J. BIOL. CHEM., (1996) vol. 271, no. 45, pp. 28154-28160.
ISSN: 0021-9258.
DOCUMENT TYPE: Journal
FILE SEGMENT: N; G; N3
LANGUAGE: English
SUMMARY LANGUAGE: English
AB The G **protein** beta subunit, G beta sub(5), is predominantly expressed in the central nervous system. In rodent brain, G beta sub(5) is expressed as a **protein** with an

apparent molecular mass of 39,000 daltons (39 kDa). We have identified an additional G beta sub(5) immunoreactive **protein** of apparent size 44 kDa in the vertebrate retina. Molecular cloning and sequencing of **polymerase** chain **reaction** products revealed that the cDNA encoding the larger species of G beta sub(5) (G beta sub(5L)) was identical to the shorter form with the addition of 126 base pairs of 5' **DNA** sequence potentially encoding an in-frame 42-amino acid extension. Sequencing of mouse G beta sub(5) genomic clones demonstrated that the 126-base pair of retinal-specific coding material is derived from a hitherto undetected 5' exon. During sucrose density gradient fractionation of bovine retinas, the 44-kDa G beta sub(5L) **protein** co-purified with rod outer segment membranes. Incubation of rod outer segment membranes with the nonhydrolyzable guanine nucleotide, GTP gamma S (guanosine 5'-3-O-(thio)triphosphate), which released the G beta subunit of transducin (G beta sub(1)), failed to remove G beta sub(5L). The 39-kDa G beta sub(5) **protein** displayed differential association with retinal and brain membranes. In the retina, G beta sub(5) was present as a soluble **protein** and was undetectable in the membrane fraction, whereas in the brain approximately 70% of G beta sub(5) was associated with cellular membranes. In transient COS-7 cell expression experiments, G beta sub(5L) formed functional G beta gamma dimers and G alpha beta gamma heterotrimers, and activated **phosphoinositide**-specific phospholipase C beta sub(2) in a manner indistinguishable from the 39-kDa G beta sub(5) **protein**. The clowning of the retinal-specific G beta sub(5L) cDNA suggests the existence of potentially novel G **protein**-mediated signaling cascades in photoreception.

L21 ANSWER 27 OF 59 MEDLINE DUPLICATE 9
ACCESSION NUMBER: 96413301 MEDLINE
DOCUMENT NUMBER: 96413301
TITLE: Biochemical and genetic defects in the **DNA**-dependent **protein** kinase in murine scid lymphocytes.
AUTHOR: Danska J S; Holland D P; Mariathasan S; Williams K M; Guidos C J
CORPORATE SOURCE: Division of Surgical Research, Hospital for Sick Children Research Institute, Toronto, Ontario, Canada.
SOURCE: MOLECULAR AND CELLULAR BIOLOGY, (1996 Oct) 16 (10) 5507-17.
Journal code: NGY. ISSN: 0270-7306.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
Searcher : Shears 308-4994

OTHER SOURCE: GENBANK-U78157; GENBANK-U78158
ENTRY MONTH: 199701
AB The scid **gene** product has been identified as the 460-kDa catalytic subunit of the **DNA**-dependent **protein** kinase (**DNA**-PKcs p460), a member of the **phosphatidylinositol 3-kinase** family. **DNA**-PK activity is undetectable in scid cells, but the molecular basis for this defect has not been identified. Here we report that expression of p460 in scid lymphocyte precursors is detectable but is reduced at least 10-fold relative to that in wild-type lymphocytes. In addition, we show that the scid mutation disturbs p460 nuclear association, presumably affecting its role in **DNA** repair pathways. To examine the molecular basis for our observations, we used a degenerate **PCR** strategy to clone the C-terminal p460 kinase domain from wild-type and scid thymocytes. Northern (**RNA**) analysis with these probes revealed normal steady-state p460 mRNA levels in scid cells, suggesting that the reduced abundance of p460 **protein** is due to a posttranscriptional defect. Sequence comparisons identified a single-base-pair alteration in the scid C-terminal p460 kinase domain, resulting in a premature stop codon. This mutation is predicted to truncate p460 by approximately 8 kDa, but it preserves the conserved motifs required for kinase activity in members of the **phosphoinositidyl** 3-kinase family. Despite a computed molecular weight alteration of less than 2%, we were able to visualize this difference by Western blot (immunoblot) analysis of wild-type and scid p460. These data demonstrate that the scid **DNA**-PKes mutation is not a null allele and suggest a molecular rationale for the well-described leakiness of the scid phenotype.

L21 ANSWER 28 OF 59 SCISEARCH COPYRIGHT 1999 ISI (R)
ACCESSION NUMBER: 97:18366 SCISEARCH
THE GENUINE ARTICLE: VY562
TITLE: Inositol trisphosphate receptor **gene** expression and hormonal regulation in osteoblast-like cell lines and primary osteoblastic cell cultures
AUTHOR: Kirkwood K L; Dziak R; Bradford P G (Reprint)
CORPORATE SOURCE: SUNY BUFFALO, DEPT PHARMACOL & TOXICOL, SCH MED & BIOMED SCI, BUFFALO, NY 14214 (Reprint); SUNY BUFFALO, DEPT PHARMACOL & TOXICOL, SCH MED & BIOMED SCI, BUFFALO, NY 14214; SUNY BUFFALO, SCH DENT MED, DEPT ORAL BIOL, BUFFALO, NY 14260; SUNY BUFFALO, CTR MOL MECHANISMS DIS & AGING, BUFFALO, NY 14260
COUNTRY OF AUTHOR: USA
SOURCE: JOURNAL OF BONE AND MINERAL RESEARCH, (DEC 1996) Vol. 11, No. 12, pp. 1889-1896.
Publisher: BLACKWELL SCIENCE INC, 238 MAIN ST,

CAMBRIDGE, MA 02142.
ISSN: 0884-0431.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE
LANGUAGE: English
REFERENCE COUNT: 53

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB The inositol trisphosphate receptor (IP3R) is an intracellular calcium channel that mediates the cellular actions of a wide variety of hormones, growth factors, and cytokines, In osteoblastic cell cultures, many bone resorbing hormones increase **phosphoinositide** turnover, inositol trisphosphate production, mobilization of intracellular calcium, and the secretion of osteoclast recruitment and activating factors, In this study, the effects of 17 beta-estradiol, 1,25-dihydroxyvitamin D-3 (1,25(OH)(2)D-3), phorbol ester, and serum on IP3R mRNA levels were evaluated in osteogenic-osteosarcoma cells and in primary osteoblastic cultures derived from neonatal rat calvaria. Type-specific reverse transcription **polymerase** chain **reaction** (RT-**PCR**) indicated that all cell types evaluated (G-292, U-2OS, Saos-2, MC3T3-E1, UMR-106, and calvarial osteoblastic cells) express IP3R mRNA type I; G-292, U-2OS, MC3T3-E1, and calvarial osteoblastic cells also express type II IP3R mRNA; and UMR-106 and the calvarial osteoblastic cells express type III IP3R mRNA. Northern blot and RT-**PCR** analyses of human G-292 osteosarcoma cells and rat calvarial osteoblastic cells showed that phorbol ester and serum increase IP3R mRNA levels, whereas 17 beta-estradiol and 1,25(OH)(2)D-3 decrease these levels. In G-292 cells, the effect of 17 beta-estradiol was not due to accelerated IP3R mRNA degradation and required continued **protein** synthesis, The results show that multiple IP3R types are expressed in osteoblasts and osteoblastic osteosarcoma cells and that this expression is regulated by 17 beta-estradiol and other osteoporotic and antiosteoporotic hormones, These findings indicate that hormonal control of IP3R expression may be relevant in the chronic regulation of osteoblast secretory activity.

L21 ANSWER 29 OF 59 EMBASE

ACCESSION NUMBER: 96110839 EMBASE
DOCUMENT NUMBER: 1996110839

TITLE: 5-HT(2B) receptor-mediated calcium release from ryanodine-sensitive intracellular stores in human pulmonary artery endothelial cells.

AUTHOR: Ullmer C.; Boddeke H.G.W.M.; Schmuck K.; Lubbert H.
CORPORATE SOURCE: Preclinical Research 386-216, Sandoz Pharma Ltd.,4002 Basel, Switzerland

SOURCE: British Journal of Pharmacology, (1996) 117/6 (1081-1088).
ISSN: 0007-1188 CODEN: BJPCBM

COUNTRY: United Kingdom
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 029 Clinical Biochemistry
030 Pharmacology
LANGUAGE: English
SUMMARY LANGUAGE: English

AB 1. We have characterized the 5-hydroxytryptamine (5-HT)-induced calcium signalling in endothelial cells from the human pulmonary artery. Using RT-**PCR** we show, that of all cloned G-**protein** coupled 5-HT receptors, these cells express only 5-HT(1D.beta.), 5-HT(2B) and little 5-HT4 receptor mRNA. 2. In endothelial cells 5-HT inhibits the formation of adenosine 3':5'-cyclic monophosphate (cyclic AMP) via 5-HT(1D.beta.) receptors but fails to activate **phosphoinositide** (PI) turnover. However, the latter pathway is strongly activated by histamine. 3. Despite the lack of detectable inositol phosphate (IP) formation in human pulmonary artery endothelial cells, 5-HT (pD2 = 5.82 .+-. 0.06, n = 6) or the selective 5-HT2 agonist, 1-(2,5-dimethoxy-4-iodophenyl)-2-aminopropane (DOI) (pD2 = 5.66 .+-. 0.03, n = 7) elicited transient calcium signals comparable to those evoked by histamine (pD2 = 6.44 .+-. 0.01, n = 7). Since 5-HT(2A) and 5-HT(2C) receptor mRNAs are not detectable in pulmonary artery endothelial cells, activation of 5-HT(2B) receptors is responsible for the transient calcium release. The calcium transients are independent of the inhibition of adenylate cyclase, since DOI does not stimulate 5-HT(1D.beta.) receptors. 4. Both, the 5-HT- and histamine-stimulated calcium signals were also observed when the cells were placed in calcium-free medium. This indicates that 5-HT triggers calcium release from intracellular stores. 5. Heparin is an inhibitor of the IP3-activated calcium release channels on the endoplasmic reticulum. Intracellular infusion of heparin through patch pipettes in voltage clamp experiments failed to block 5-HT-induced calcium signals, whereas it abolished the histamine response. This supports the conclusion that the 5-HT-induced calcium release is independent of IP3 formation. 6. Unlike the histamine response, the 5-HT response was sensitive to micromolar concentrations of ryanodine and, to a lesser extent, ruthenium red. This implies that 5-HT(2B) receptors trigger calcium release from a ryanodine-sensitive calcium pool. 7. It has been postulated that cyclic ADP-ribose (cADPR) is a soluble second messenger which activates ryanodine receptors. However, calcium signals similar to the 5-HT response could not be elicited by intracellular infusion with cADPR. Furthermore, the subsequent application of 5-HT or DOI elicited a calcium signal that was not affected by the above pretreatment. 8. We conclude that human 5-HT(2B) receptors stimulate calcium release from intracellular stores through a novel pathway, which involves activation of ryanodine receptors, and is independent of PI-hydrolysis and cADPR.

L21 ANSWER 30 OF 59 MEDLINE DUPLICATE 10

ACCESSION NUMBER: 97017223 MEDLINE
DOCUMENT NUMBER: 97017223
TITLE: Identification, molecular cloning, and distribution of a short variant of the 5-hydroxytryptamine2C receptor produced by alternative splicing.
AUTHOR: Canton H; Emeson R B; Barker E L; Backstrom J R; Lu J T; Chang M S; Sanders-Bush E
CORPORATE SOURCE: Department of Pharmacology, Vanderbilt University School of Medicine, Nashvile, Tennessee 37232, USA.
CONTRACT NUMBER: MH34007 (NIMH)
SOURCE: MOLECULAR PHARMACOLOGY, (1996 Oct) 50 (4) 799-807. Journal code: NGR. ISSN: 0026-895X.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
OTHER SOURCE: GENBANK-U35315
ENTRY MONTH: 199701
ENTRY WEEK: 19970104

AB The actions of the neurotransmitter 5-hydroxytryptamine (5-HT) (serotonin) are mediated by multiple receptor subtypes. One of the prominent serotonin receptors in the brain is the 5-HT2C receptor (5-HT2C-R). We report the occurrence of a second 5-HT2C-R transcript, first identified using S1 nuclease protection of total **RNA** isolated from the choroid plexus. Analyses of the distribution of these two **RNAs** revealed that the short form is expressed in the same structures as the 5-HT2C-R mRNA, including choroid plexus, striatum, hippocampus, hypothalamus, olfactory tubercles, and spinal cord. Cloning and sequence analyses revealed a second cDNA with a 95-nt deletion in the region coding for the putative second intracellular loop and the fourth transmembrane domain of the 5-HT2C-R. This deletion leads to a frameshift in the coding sequence and the introduction of a premature stop codon. The predicted truncated **protein** (5-HT2C-tr) contains 172 amino acids, with 153 residues at the amino terminus, identical to the 5-HT2C-R, and 19 carboxyl-terminal amino acids that are unique. Although antibodies specific to the 5-HT2C-tr **protein** showed that the truncated form is expressed in a transfected fibroblast cell model system, there was no serotonergic ligand binding activity or **phosphoinositide** hydrolysis. Analyses of the 5-HT2C-R **gene** revealed that the two transcripts arise from a single **gene** by differential splicing using alternative donor sites and a common 3'-splice acceptor. **Polymerase** chain **reaction** amplification of mouse and human brain cDNAs demonstrated the occurrence of the same splicing patterns in these species. Although this study demonstrates tissue-specific expression of two 5-HT2C mRNA splice variants in rat, mouse, and human, the significance of

the truncated form in these three species remains to be established.

L21 ANSWER 31 OF 59 MEDLINE
ACCESSION NUMBER: 96296280 MEDLINE
DOCUMENT NUMBER: 96296280
TITLE: Functional characterisation of an ovine endometrial oxytocin receptor cDNA transiently expressed in Cos-7 cells.
AUTHOR: Riley P R; Abayasekara D R; Stewart H J; Flint A P
CORPORATE SOURCE: Department of Physiology, Royal Free Hospital School of Medicine, London, UK.
SOURCE: JOURNAL OF ENDOCRINOLOGY, (1996 Jun) 149 (3) 389-96. Journal code: I1J. ISSN: 0022-0795.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199610

AB The entire coding region of an ovine endometrial oxytocin receptor (OTR) cDNA was generated by **PCR**, subcloned into the SV40 major late promoter expression vector pSVLJ and transiently expressed in Cos-7 cells. A specific OTR antagonist, 125I-labelled d(CH2)5 [Tyr(Me)2,Thr4,Tyr-NH2(9)]-vasotocin (OTA), was used to describe the binding kinetics of the expressed receptor which had a Kd of 4.5 nM and Bmax of 2.4 nM/mg **protein** (6.8 x 10(5) receptor molecules/transfected cell). The functional properties of the expressed OTR were determined by measuring oxytocin-induced **phosphoinositide** (PI) hydrolysis. Oxytocin increased PI turnover in OTR transfected cells fourfold in excess of residual endogenous activity, and stimulated phospholipase C (PLC) activity in a dose- and time-dependent manner, confirming that the expressed OTR cDNA was functional. Arginine vasopressin also stimulated PI turnover in a dose-dependent manner; thresholds of responses to oxytocin and arginine vasopressin were 10(-9) M and 10(-7) M respectively. OTA did not increase PI turnover and competitively inhibited the oxytocin-induced response. Direct activation of the pathway by aluminium fluoride and guanosine (3'-O-thio)-triphosphate (GTP gamma S) confirmed that the OTR was G-**protein** linked. Co-incubation of GTP gamma S with oxytocin shifted the PI-response threshold from 10(-7) M to 10(-9) M and significantly increased the level of response, suggesting that maximum PI turnover was agonist-dependent. The G-**protein** involved in mediating the signal transduction pathway was pertussis toxin-insensitive and, therefore, probably a member of the Gq subfamily. The PLC inhibitor, U73122, had no effect on oxytocin-induced PI turnover, consistent with the response in endometrial tissue. These data suggest that the signalling pathway mediated by expressed OTR is similar to that attributed to OTR occupancy in ovine endometrium.

L21 ANSWER 32 OF 59 LIFESCI COPYRIGHT 1999 CSA
ACCESSION NUMBER: 96:101243 LIFESCI
TITLE: Deletion of Gly super(723) in the insulin receptor substrate-1 of a patient with noninsulin-dependent diabetes mellitus
AUTHOR: Esposito, D.L.; Mammarella, S.; Ranieri, A.; Loggia, F.D.; Capani, F.; Consoli, A.; Mariani-Costantini, R.; Caramia, F.G.; Cama, A.*; Battista, P.
CORPORATE SOURCE: Ist. Patol. Umana e Med. Soc., Univ. Chieti, Italy, "Gabriele D'Annunzio," 66013 Chieti, Italy
SOURCE: HUM. MUTAT., (1996) vol. 7, no. 4, pp. 364-366.
ISSN: 1059-7794.
DOCUMENT TYPE: Journal
FILE SEGMENT: G
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Insulin receptor substrate-1 (IRS-1), the major substrate of the insulin and IGF-1 receptor tyrosine kinases, is a multisite docking **protein** that contains 21 potential tyrosine phosphorylation sites, including six in Tyr-Met-Xaa-Met (YMXM) motifs, three in Tyr-Xaa-Xaa-Met (YXXM) motifs, and 12 in other hydrophobic motifs. Phosphorylated tyrosine residues included in these motifs appear to be crucial for the interaction of IRS-1 with src homology 2 (SH2) domains, present in downstream signalling molecules such as GRB-2, the 85K regulatory subunit of **phosphatidylinositol 3-kinase** (p85 alpha), SHPTP2 (Syp), and Nck. IRS-1 plays a key role in mediating insulin action, and IRS-1 mutations have been implicated recently in the pathogenesis of noninsulin dependent diabetes mellitus (NIDDM). Using genomic **DNA** as a template, we analyzed by SSCP the region of the IRS-1 **gene** spanning codons 624-755 (amino acid numbering according to Araki et al., 1993) in 63 unrelated NIDDM patients and 47 control subjects. The analysis revealed the presence of a unique conformer in one NIDDM patient (Fig. 1). Direct sequencing of the **PCR** product, using the primers designed for nested amplification, revealed the presence of an in-frame deletion, in the context of two GGT direct repeats located at codons 722 and 723 (data not shown).

L21 ANSWER 33 OF 59 LIFESCI COPYRIGHT 1999 CSA
ACCESSION NUMBER: 97:12268 LIFESCI
TITLE: Alterations in the level of phosphotyrosine signal transduction constituents in human parotid tumors
AUTHOR: Bu, Rongfa; Purushotham, K.R.; Kerr, M.; Tao, Zeng; Jonsson, R.; Olofsson, J.; Humphreys-Beher, M.G.*
CORPORATE SOURCE: Dep. Oral Biol., P.O. Box 100424, Univ. Florida, Gainesville, FL 32610, USA
SOURCE: PROC. SOC. EXP. BIOL. MED., (1996) vol. 211, no. 3, pp. 257-264.
ISSN: 0037-9727.

DOCUMENT TYPE: Journal
FILE SEGMENT: N
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Human parotid tumors were evaluated for the activation of the phosphotyrosine signaling pathway by Western blot, enzyme activity assay, and reverse transcriptase-**polymerase** chain **reaction**. Warthin's tumor and mucoepidermoid carcinomas had the greatest level of tyrosine phosphorylated **proteins** identified in plasma membrane fractions. These tumors, along with pleomorphic adenocarcinoma, showed high levels of membrane expression of the tyrosine kinase receptor, c-erbB-2, and **phosphatidylinositol-3-kinase**.
Expression of the epidermal growth factor receptor was confined to normal tissue. The level of mRNA for c-erb was elevated only in mucoepidermoid carcinomas. Messenger **RNA** levels for ras were unchanged from control levels in all tumors, while the level of src mRNA was higher in the tumor samples than the normal parotid tissue. The activities of several signal transduction kinases, including **protein** kinase A and C were elevated in tumor tissue (7.7- to 18.9- and 0.4- to 3.7-fold higher, respectively), relative to surrounding normal tissue. While the level of glandular amylase was reduced (22%-0% of normal levels) in the tumor tissue, epidermal growth factor (EGF) and transforming growth factor- alpha (TGF alpha) content was dramatically higher in the neoplastic tissue (10- to 170-fold and 4.6- to 6.0-fold, respectively). These results suggest that with the presence of elevated levels of EGF, TGF alpha , and the oncoprotein receptor c-erbB-2 in the membrane of parotid tumors, cell proliferation and activation of the phosphotyrosine signal transduction pathway may involve autocrine stimulation through the expression of high levels of growth factor and receptor in the same tissue.

L21 ANSWER 34 OF 59 MEDLINE DUPLICATE 11
ACCESSION NUMBER: 97101871 MEDLINE
DOCUMENT NUMBER: 97101871
TITLE: Molecular cloning of the Toxoplasma gondii sag4 **gene** encoding an 18 kDa bradyzoite specific surface **protein**.
AUTHOR: Odberg-Ferragut C; Soete M; Engels A; Samyn B; Loyens A; Van Beeumen J; Camus D; Dubremetz J F
CORPORATE SOURCE: INSERM U42, Villeneuve d'Ascq, France..
101473.2353@CompuServe.COM
SOURCE: MOLECULAR AND BIOCHEMICAL PARASITOLOGY, (1996 Nov 25) 82 (2) 237-44.
Journal code: NOR. ISSN: 0166-6851.
PUB. COUNTRY: Netherlands
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English

FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-Z69373
ENTRY MONTH: 199707
ENTRY WEEK: 19970703

AB An 18 kDa bradyzoite specific surface **protein** of Toxoplasma gondii (T. gondii) has been purified by affinity chromatography with a specific monoclonal antibody using parasites grown in vitro under conditions inducing the biosynthesis of bradyzoite specific **proteins**. N-terminal and internal amino acid sequences obtained by microsequencing enabled us to design degenerate oligonucleotides. A fragment of 187 bp was amplified by **polymerase** chain **reaction** (**PCR**). It was used as a probe to clone a 4 kb-Bam HI fragment encompassing the **gene** encoding the 18 kDa **protein** . Nucleotide sequence analysis revealed a single open reading frame of 516 nucleotides encoding a 172 amino acid **protein**. The deduced amino acid sequence matched perfectly the **peptides** microsequenced from the native **protein**. The N-terminal hydrophobic region was found to possess the characteristics of a signal **peptide** of 27 amino acids. The hydrophobic C-terminal part could represent a signal for a glycan-**phosphoinositide** anchor. The full-length cDNA was also isolated and both the 5' and 3' untranslated regions were determined. Reverse transcriptase-**PCR** (RT-**PCR**) using p18-specific primers showed a stage-specific expression of this **gene**. Comparison of the **nucleic** acid sequence and the predicted amino acid sequence with databases did not reveal significant homology with known **genes** or **proteins**. This **gene** is proposed to be named sag4, according to the existing T. gondii nomenclature.

L21 ANSWER 35 OF 59 MEDLINE
ACCESSION NUMBER: 96438850 MEDLINE
DOCUMENT NUMBER: 96438850
TITLE: The Friedreich's ataxia **gene** encodes a novel phosphatidylinositol-4- phosphate 5-kinase [see comments].
COMMENT: Comment in: Nat Genet 1997 Apr;15(4):337-8
AUTHOR: Carvajal J J; Pook M A; dos Santos M; Doudney K; Hillermann R; Minogue S; Williamson R; Hsuan J J; Chamberlain S
CORPORATE SOURCE: Department of Biochemistry and Molecular Genetics, Imperial College School of Medicine at St. Mary's, London, UK.
SOURCE: NATURE GENETICS, (1996 Oct) 14 (2) 157-62. Journal code: BRO. ISSN: 1061-4036.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English

FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-U14957; GENBANK-U60871; GENBANK-X92493
ENTRY MONTH: 199701
AB The STM7 **gene** on chromosome 9 was recently 'excluded' as a candidate for Friedreich's ataxia following the identification of an expanded intronic GAA triplet repeat in the adjacent **gene**, X25, in patients with the disease. Using RT-**PCR**, northern and sequence analyses, we now demonstrate that X25 comprises part of the STM7 **gene**, contributing to at least four splice variants, and report the identification of new coding sequences. Functional analysis of the STM7 recombinant **protein** corresponding to the reported 2.7-kilobase transcript has demonstrated PtdInsP 5-kinase activity, supporting the idea that the disease is caused by a defect in the **phosphoinositide** pathway, possibly affecting vesicular trafficking or synaptic transmission.

L21 ANSWER 36 OF 59 BIOSIS COPYRIGHT 1999 BIOSIS DUPLICATE 12
ACCESSION NUMBER: 1996:414784 BIOSIS
DOCUMENT NUMBER: PREV199699137140
TITLE: Acute regulation by insulin in phosphatidylinositol-3-kinase, Rad, Glut 4, and lipoprotein lipase mRNA levels in human muscle.
AUTHOR(S): Laville, M. (1); Auboeuf, D.; Khalfallah, Y.; Vega, N.; Riou, J. P.; Vidal, H.
CORPORATE SOURCE: (1) INSERM U449, Fac. Med. Alexis Carrel, Rue G. Paradin, 69373 Lyon Cedex 08 France
SOURCE: Journal of Clinical Investigation, (1996) Vol. 98, No. 1, pp. 43-49.
ISSN: 0021-9738.
DOCUMENT TYPE: Article
LANGUAGE: English
AB We have investigated the acute regulation by insulin of the mRNA levels of nine **genes** involved in insulin action, in muscle biopsies obtained before and at the end of a 3-h euglycemic hyperinsulinemic clamp. Using reverse transcription-competitive **PCR**, we have measured the mRNAs encoding the two insulin receptor variants, the insulin receptor substrate-1, the p85a subunit of **phosphatidylinositol-3-kinase**, Ras associated to diabetes (Rad), the glucose transporter Glut 4, glycogen synthase, 6-phosphofructo-1-kinase, lipoprotein lipase, and the hormone-sensitive lipase. Insulin infusion induced a significant increase in the mRNA level of Glut 4 (+56 +- 13%), Rad (+96 +- 25%), the p85-alpha subunit of **phosphatidylinositol-3-kinase** (+92 +- 18%) and a decrease in the lipoprotein lipase mRNA level (-49 +- 5%), while the abundance of the other mRNAs was unaffected. The relative expression of the two insulin receptor variants was not modified. These results demonstrate an acute coordinated regulation by insulin of the expression of **genes**

coding key **proteins** involved in its action in human skeletal muscle and suggest that Rad and the p85-alpha regulatory subunit of **phosphatidylinositol-3-kinase** can be added to the list of the **genes** controlled by insulin.

L21 ANSWER 37 OF 59 MEDLINE DUPLICATE 13
ACCESSION NUMBER: 96256846 MEDLINE
DOCUMENT NUMBER: 96256846
TITLE: Ataxia-telangiectasia: mutations in ATM cDNA detected by **protein**-truncation screening.
AUTHOR: Telatar M; Wang Z; Udar N; Liang T; Bernatowska-Matuszkiewicz E; Lavin M; Shiloh Y; Concannon P; Good R A; Gatti R A
CORPORATE SOURCE: Department of Pathology, School of Medicine, University of California, Los Angeles, USA.
CONTRACT NUMBER: CA57569 (NCI)
SOURCE: AMERICAN JOURNAL OF HUMAN GENETICS, (1996 Jul) 59 (1) 40-4.
Journal code: 3IM. ISSN: 0002-9297.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199610
AB We have examined the distal half of the ataxia-telangiectasia (A-T) **gene** transcript for truncation mutations in 48 A-T affecteds. We found 21 mutations; 4 of the mutations were seen in more than one individual. Genotyping of the individuals sharing mutations, by using nearby microsatellite markers, established that three of the four groups shared common haplotypes, indicating that these were probably founder effects, not public mutations. The one public mutation was found in two American families, one of Ashkenazi Jewish background and the other not. Most truncations deleted the **PI3-kinase** domain, although some exceptions to this were found in patients with typical A-T phenotypes. All patients not previously known to be consanguineous were found to be compound heterozygotes when mutations could be identified--that is, normal and abnormal **protein** segments were seen on SDS-PAGE gels. All 48 patients gave RT-**PCR** products, indicating the presence of relatively stable mRNAs despite their mutations. These results suggest that few public mutations or hot spots can be expected in the A-T **gene** and that epidemiological studies of A-T carrier status and associated health risks will have to be designed around populations with frequent founder-effect mutations, despite the obvious limitations of this approach.

L21 ANSWER 38 OF 59 LIFESCI COPYRIGHT 1999 CSA DUPLICATE 14
ACCESSION NUMBER: 96:36797 LIFESCI

TITLE: Molecular cloning, splice variants, expression, and purification of phospholipase C- delta 4
AUTHOR: Lee, Sang Bong; Rhee, Sue Goo
CORPORATE SOURCE: Natl. Inst. Health, 9000 Rockville Pike, Bldg. 3, Rm. 122, 3 CENER DR MSC 0340, Bethesda, MD 20892-0340, USA
SOURCE: J. BIOL. CHEM., (1996) vol. 271, no. 1, pp. 25-31. ISSN: 0021-9258.
DOCUMENT TYPE: Journal
FILE SEGMENT: N; G
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Complementary **DNAs** encoding a previously unidentified **phosphoinositide**-specific phospholipase C (PLC) isozyme were cloned from a rat brain cDNA library by the **polymerase** chain **reaction** with degenerate oligonucleotide primers based on sequences common to three known delta -type PLC isozymes. The encoded **polypeptide** contains 772 amino acids (calculated molecular mass, 88,966 daltons) and is similar in primary structure to delta -type PLC isozymes, with overall sequence identities of 45% to PLC- delta 1, 72% to PLC- delta 2, and 47% to PLC- delta 3. Thus, the new PLC isozyme was named PLC- delta 4. Recombinant PLC- delta 4 was purified from extracts of HeLa cells that had been infected with vaccinia virus containing the corresponding cDNA. The purified **protein** exhibited an apparent molecular mass of 90 kDa on SDS-polyacrylamide gels. The specific activity of PLC- delta 4 and its dependence on Ca super(2+) were similar to those of PLC- delta 1. The distribution of PLC- delta 4 in 16 different rat tissues was studied by immunoblot analysis with PLC- delta 4-specific antibodies of fractions obtained after an enzyme-enrichment procedure. The 90-kDa immunoreactive **protein** was detected unambiguously in only eight tissues and was present at concentrations that were low compared to those of other major PLC isozymes. A 93-kDa immunoreactive **protein** was also prominent in testis but was not detected in the other seven positive tissues. The 93-kDa enzyme appears to be derived from a splice variant of the mRNA that encodes the 90-kDa PLC- delta 4 and contains an additional 32 amino acids between the X and Y catalytic domains. Splice variants have not previously been detected for delta -type PLC isozymes.

L21 ANSWER 39 OF 59 MEDLINE DUPLICATE 15
ACCESSION NUMBER: 96102072 MEDLINE
DOCUMENT NUMBER: 96102072
TITLE: Identification of two isoforms of mouse neuropeptide Y-Y1 receptor generated by alternative splicing. Isolation, genomic structure, and functional expression of the receptors.
AUTHOR: Nakamura M; Sakanaka C; Aoki Y; Ogasawara H; Tsuji T;

Kodama H; Matsumoto T; Shimizu T; Noma M
CORPORATE SOURCE: Life Science Research Laboratory, Japan Tobacco Inc., Kanagawa, Japan.
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (1995 Dec 15) 270 (50) 30102-10.
Journal code: HIV. ISSN: 0021-9258.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
OTHER SOURCE: GENBANK-D63818; GENBANK-D63819
ENTRY MONTH: 199604

AB Two cDNA clones homologous with human neuropeptide (NP) Y-Y1 receptor have been isolated from a mouse bone marrow cDNA library. One was thought to be the cognate of the human NPY-Y1 receptor, termed Y1 alpha receptor, and the other form, termed Y1 beta receptor, differed from the Y1 alpha receptor in the seventh transmembrane domain and C-terminal tail. Analysis of the mouse genomic **DNA** showed that both receptors originated from a single **gene**. The different **peptide** sequences of the Y1 beta receptor were encoded by separate exons, hence, these receptors were generated by differential **RNA** splicing. High affinity binding of [125I]NPY to each receptor expressed in Chinese hamster ovary (CHO) cells and sequestration of [125I]NPY after binding to each receptor were observed. In the CHO cells expressing the Y1 alpha receptor, intracellular Ca2+ increase, inhibition of forskolin-induced cAMP accumulation, and mitogen-activated **protein** kinase (MAPK) activation were observed by stimulation of NPY, and these responses were abolished by pretreatment with pertussis toxin. Since wortmannin completely inhibited NPY-elicited MAPK activation, we speculate that wortmannin-sensitive signaling molecule(s) such as **phosphoinositide** 3-kinase may lie between pertussis toxin-sensitive G-**protein** and MAPK. In contrast, these intracellular signals were not detected in CHO cells expressing the Y1 beta receptor. Northern blots and reverse transcriptase-**polymerase** chain **reaction** analyses indicated that the Y1 alpha receptor was highly expressed in the brain, heart, kidney, spleen, skeletal muscle, and lung, whereas the Y1 beta receptor mRNA was not detected in these tissues. However, the Y1 beta receptor was expressed in mouse embryonic developmental stage (7 and 11 days), bone marrow cells and several hematopoietic cell lines. These results suggest that the Y1 beta receptor is an embryonic and a bone marrow form of the NPY-Y1 receptor, which decreases in the expression during development and differentiation.

L21 ANSWER 40 OF 59 MEDLINE
ACCESSION NUMBER: 96036069 MEDLINE
DOCUMENT NUMBER: 96036069

TITLE: Grb-IR: a SH2-domain-containing **protein** that binds to the insulin receptor and inhibits its function.
AUTHOR: Liu F; Roth R A
CORPORATE SOURCE: Department of Molecular Pharmacology, Stanford University School of Medicine, CA 94305, USA.
CONTRACT NUMBER: DK41765 (NIDDK)
DK34962 (NIDDK)
SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (1995 Oct 24) 92 (22) 10287-91.
Journal code: PV3. ISSN: 0027-8424.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
OTHER SOURCE: GENBANK-U34355
ENTRY MONTH: 199602

AB To identify potential signaling molecules involved in mediating insulin-induced biological responses, a yeast two-hybrid screen was performed with the cytoplasmic domain of the human insulin receptor (IR) as bait to trap high-affinity interacting **proteins** encoded by human liver or HeLa cDNA libraries. A SH2-domain-containing **protein** was identified that binds with high affinity in vitro to the autophosphorylated IR. The mRNA for this **protein** was found by Northern blot analyses to be highest in skeletal muscle and was also detected in fat by **PCR**. To study the role of this **protein** in insulin signaling, a full-length cDNA encoding this **protein** (called Grb-IR) was isolated and stably expressed in Chinese hamster ovary cells overexpressing the human IR. Insulin treatment of these cells resulted in the in situ formation of a complex of the IR and the 60-kDa Grb-IR. Although almost 75% of the Grb-IR **protein** was bound to the IR, it was only weakly tyrosine-phosphorylated. The formation of this complex appeared to inhibit the insulin-induced increase in tyrosine phosphorylation of two endogenous substrates, a 60-kDa GTPase-activating-**protein**-associated **protein** and, to a lesser extent, IR substrate 1. The subsequent association of this latter **protein** with **phosphatidylinositol 3-kinase** also appeared to be inhibited. These findings raise the possibility that Grb-IR is a SH2-domain-containing **protein** that directly complexes with the IR and serves to inhibit signaling or redirect the IR signaling pathway.

L21 ANSWER 41 OF 59 MEDLINE
ACCESSION NUMBER: 95237103 MEDLINE
DOCUMENT NUMBER: 95237103
TITLE: Signaling properties of mouse and human

corticotropin-releasing factor (CRF) receptors: decreased coupling efficiency of human type II CRF receptor [see comments].

COMMENT: Comment in: Endocrinology 1995 May;136(5):1819-20

AUTHOR: Xiong Y; Xie L Y; Abou-Samra A B
CORPORATE SOURCE: Endocrine Unit, Massachusetts General Hospital, Boston 02114, USA.

CONTRACT NUMBER: NIDDK 45020
SOURCE: ENDOCRINOLOGY, (1995 May) 136 (5) 1828-34.
Journal code: EGZ. ISSN: 0013-7227.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals; Cancer Journals
ENTRY MONTH: 199507

AB CRF is the primary neuroregulator of the function of the hypothalamic-pituitary-adrenal axis. We have recently cloned a mouse CRF receptor (mCRF-R) complementary **DNA** (cDNA) from an AtT-20 cell cDNA library by **polymerase** chain **reaction**. To compare the functions of mCRF-R to those of the human type I and type II CRF receptors (hCRF-RI and hCRF-RII), cDNAs were cloned into the expression vector pcDNA1 and transfected into COS-7 cells. CRF binding and CRF-stimulated cAMP accumulation as well as **phosphoinositide** hydrolysis were measured. Scatchard analysis of the binding of 125I-labeled [Tyr0]r/hCRF ([125I]CRF) to COS-7 cells expressing mCRF-R and hCRF-RI cDNAs revealed the same apparent Kd (9 nM). In contrast, the apparent binding Kd for hCRF-RII was 20 nM CRF. Maximal stimulatory concentrations (1 microM) of rat/human CRF-(1-41) (r/hCRF) increased cAMP accumulation in COS-7 cells transfected with mCRF-R, hCRF-RI, and hCRF-RII cDNA plasmid (10 micrograms each) from basal values of 8-19 pmol/10(5) cells.15 min to 84 +/- 10, 87 +/- 16, and 45 +/- 16 pmol/10(5) cells.15 min, respectively. The EC50 values of r/hCRF-stimulated cAMP accumulation in COS-7 cells expressing mCRF-R and hCRF-RI cDNAs were similar at 0.4 +/- 0.2 and 0.7 +/- 0.2 nM, respectively. Conversely, the EC50 of r/hCRF-stimulated cAMP accumulation in hCRF-RII-transfected COS-7 cells was 47.5 +/- 18.9 nM. As the level of expression of hCRF-RII was lower than that of hCRF-RI, we compared r/hCRF-stimulated cAMP accumulation in COS-7 cells expressing low and high levels of hCRF-RI. The EC50 for r/hCRF-stimulated cAMP accumulation in COS-7 cells transfected with hCRF-RI did not change when receptor expression was varied by a factor of 1- to 8.4-fold. In contrast, the EC50 for r/hCRF-stimulated cAMP accumulation mediated by hCRF-RII was at least 100-fold higher than that mediated by the hCRF-RI in COS-7 cells, which suggests poor coupling between hCRF-RII and adenylate cyclase. Inositol phosphate (IP) levels were also determined in mCRF-R, hCRF-RI, and hCRF-RII cDNA-transfected COS-7 cells

stimulated with increasing concentrations of r/hCRF. r/hCRF-stimulated IPs accumulation was dose dependent in COS-7 cells expressing mCRF-R and hCRF-RI using 100 and 1000 nM r/hCRF. Concentrations of 10 (or less) nM r/hCRF had no effect on IP generation. hCRF-RII did not mediate stimulation of IP even at 1000 nM r/hCRF. (ABSTRACT TRUNCATED AT 400 WORDS)

L21 ANSWER 42 OF 59 MEDLINE
ACCESSION NUMBER: 95385853 MEDLINE
DOCUMENT NUMBER: 95385853
TITLE: Frequency of mutations of insulin receptor **gene** in Japanese patients with NIDDM.
AUTHOR: Kan M; Kanai F; Iida M; Jinnouchi H; Todaka M; Imanaka T; Ito K; Nishioka Y; Ohnishi T; Kamohara S; et al
CORPORATE SOURCE: Department of Enzyme Genetics, Institute for Enzyme Research, University of Tokushima, Japan.
SOURCE: DIABETES, (1995 Sep) 44 (9) 1081-6.
Journal code: E8X. ISSN: 0012-1797.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199512
AB To examine the prevalence of abnormalities in the insulin receptor structure **gene** in Japanese with non-insulin-dependent diabetes mellitus (NIDDM), a population of 51 patients with NIDDM was screened for mutations in this **gene**. Patient genomic **DNAs** of both alleles corresponding to 22 exons of the **gene** were amplified by **polymerase** chain **reaction** (**PCR**). The **PCR** products on pUC19 were sequenced. Three patients with heterozygous missense mutation Thr831-->Ala831 in exon 13 and one patient with heterozygous missense mutation Tyr1334-->Cys1334 in exon 22 of the beta-subunits were identified. Linkage analysis of one of the families plus statistical studies showed that the mutation Thr831-->Ala831 is possibly responsible for the onset of NIDDM. In COS cells transiently expressing both mutant receptor cDNAs and a cDNA of a M(r) 85,000 regulatory subunit of **phosphatidylinositol 3-kinase** (PI 3-kinase), the mutation Tyr1334-->Cys1334 impaired binding of the receptor with the M(r) 85,000 subunit of PI 3-kinase, but linkage analysis of the family showed that the mutation did not cosegregate with NIDDM in the pedigree. Therefore, one missense mutation (Thr831-->Ala831) in the insulin receptor, as found in three patients, is possibly involved in the etiology of a subset of the 51 NIDDM patients.

L21 ANSWER 43 OF 59 SCISEARCH COPYRIGHT 1999 ISI (R)
Searcher : Shears 308-4994

ACCESSION NUMBER: 95:825854 SCISEARCH
THE GENUINE ARTICLE: TG288
TITLE: MOLECULAR-CLONING AND EXPRESSION OF MULTIPLE ISOFORMS OF HUMAN PROSTAGLANDIN-E RECEPTOR EP(3) SUBTYPE GENERATED BY ALTERNATIVE MESSENGER-RNA SPLICING - MULTIPLE 2ND-MESSENGER SYSTEMS AND TISSUE-SPECIFIC DISTRIBUTIONS
AUTHOR: KOTANI M; TANAKA I (Reprint); OGAWA Y; USUI T; MORI K; ICHIKAWA A; NARUMIYA S; YOSHIMI T; NAKAO K
CORPORATE SOURCE: KYOTO UNIV, FAC MED, DEPT MED, DIV 2, SAKYO KU, 54 SHOGOIN KAWAHARA CHO, KYOTO 606, JAPAN (Reprint); KYOTO UNIV, FAC MED, DEPT MED, DIV 2, SAKYO KU, KYOTO 606, JAPAN; KYOTO UNIV, FAC MED, DEPT PHARMACOL, SAKYO KU, KYOTO 606, JAPAN; KYOTO UNIV, FAC PHARMACEUT SCI, DEPT PHYSIOL CHEM, SAKYO KU, KYOTO 606, JAPAN; HAMAMATSU UNIV, SCH MED, DEPT INTERNAL MED 2, HAMAMATSU, SHIZUOKA 43131, JAPAN
COUNTRY OF AUTHOR: JAPAN
SOURCE: MOLECULAR PHARMACOLOGY, (NOV 1995) Vol. 48, No. 5, pp. 869-879.
ISSN: 0026-895X.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE
LANGUAGE: ENGLISH
REFERENCE COUNT: 41
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB Five distinct cDNA clones encoding four different isoforms of human prostaglandin (PG) E receptor EP(3) subtype were isolated from a human kidney cDNA library. Two cDNA clones differed only in their 3'-untranslated regions. The four isoforms, tentatively named EP(3-I), EP(3-II), EP(3-III), and EP(3-IV), which are generated by alternative mRNA splicing, had identical amino acid sequences except for their different carboxyl-terminal tails. Transfection experiments revealed that all the four isoforms show high binding affinities to PGE(2), PGE(1), and M&B28767, an EP(3)-specific agonist, whereas their downstream signaling pathways are divergent. M&B28767 increased cAMP concentrations in cells expressing EP(3-II) and EP(3-IV), whereas it inhibited forskolin-induced cAMP accumulations in cells expressing all EP(3) isoforms. M&B28767 also stimulated **phosphoinositide** turnover in cells expressing EP(3-I) and EP(3-II). Northern blot analysis revealed that the EP(3) **gene** is expressed in a wide variety of human tissues. The human EP(3) mRNA was present most abundantly in the kidney, pancreas, and uterus. A substantial expression was also detected in the heart, liver, skeletal muscle, small intestine, colon, prostate, ovary, and testis. Furthermore, reverse transcription-**polymerase** chain **reaction** analysis demonstrated tissue-specific expressions of the five different EP(3) mRNA species. The present study suggests the presence of the multiple

systems of PGE(2)/EP(3) isoforms and leads to the better understanding of its physiological and pathophysiological implications in humans.

L21 ANSWER 44 OF 59 EMBASE COPYRIGHT 1999 ELSEVIER SCI. B.V.
ACCESSION NUMBER: 95351072 EMBASE
DOCUMENT NUMBER: 1995351072
TITLE: Cloning of the rat ErbB3 cDNA and characterization of the recombinant **protein**.
AUTHOR: Hellyer N.J.; Kim H.-H.; Greaves C.H.; Sierke S.L.; Koland J.G.
CORPORATE SOURCE: Department of Pharmacology, University of Iowa, College of Medicine,Iowa City, IA 52242-1109, United States
SOURCE: Gene, (1995) 165/2 (279-284).
ISSN: 0378-1119 CODEN: GENED6
COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 022 Human Genetics
029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Three cDNA fragments that encoded all but the extreme N terminus of the rat ErbB3 **protein** were cloned by low-stringency screening of a rat liver cDNA library with a human ERBB3 probe. The remaining 5'-end of the cDNA was generated by a reverse transcription-**polymerase** chain **reaction** method, and a single full-length rat ErbB3 cDNA was assembled. A comparison of the deduced amino acid (aa) sequences of human and rat ErbB3 was made, and the effects of certain aa substitutions in the putative **protein** tyrosine kinase domain were considered. The rat ERbB3 cDNA was subsequently expressed in cultured NIH-3T3 mouse fibroblasts, in which a high level of approx. 180-kDa recombinant ErbB3 (re-ErbB3) was generated. The rat re-ErbB3 produced in transfected fibroblasts was responsive to the **polypeptide**, heregulin, a known ligand for ErbB3. Challenge of transfected fibroblasts with heregulin stimulated the phosphorylation of rat re-ErbB3 on Tyr residues and promoted its association with the p85 subunit of **phosphatidylinositol 3-kinase** . Together, these results indicate that a fully functional rat ErbB3 cDNA has been isolated, and that fibroblast cells expressing this cDNA will be suitable for investigations of the signal transduction mechanism of ErbB3.

L21 ANSWER 45 OF 59 MEDLINE DUPLICATE 16
ACCESSION NUMBER: 96004802 MEDLINE
DOCUMENT NUMBER: 96004802
TITLE: cDNA encoding a functional feline liver/bone/kidney-type alkaline phosphatase.

AUTHOR: Ghosh A K; Mullins J I
CORPORATE SOURCE: Department of Microbiology and Immunology, Stanford University School of Medicine, California 94305-5402, USA.
CONTRACT NUMBER: CA43216 (NCI)
CA59046 (NCI)
SOURCE: ARCHIVES OF BIOCHEMISTRY AND BIOPHYSICS, (1995 Sep 10) 322 (1) 240-9.
Journal code: 6SK. ISSN: 0003-9861.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
OTHER SOURCE: GENBANK-U31569
ENTRY MONTH: 199601
AB Feline alkaline phosphatase (FeALP) was copurified with the putative 70-kDa feline leukemia virus subgroup-A (FeLV-A) receptor **protein** from feline T-lymphocyte cells (FeT) by two-dimensional gel electrophoresis. The sequence of the N-terminal 17 amino acids and five other internal tryptic **peptides** revealed that it is homologous to the liver/bone/kidney (L/B/K)-type alkaline phosphatase of other mammalian species. Corresponding oligonucleotides were synthesized and used for amplification of a 1.2-kb segment of the FeALP **gene** by **polymerase** chain **reaction**, using phage **DNA** from a FeT cell cDNA library as template. The 1.2-kb FeALP **gene** fragment generated was then used as a probe to isolate a 2127-bp L/B/K-type FeALP cDNA clone from the same library harboring a large, intact open reading frame. This cDNA possessed an open reading frame encoding a 524-amino-acid **protein** including a putative signal **peptide** of 17 amino acids followed by 14-amino-acid residues identical to the N-terminal sequence determined from the purified **protein**. Sequences closely related to five tryptic **peptides** from the purified **protein** were also contained within the cDNA-encoded **protein**. Homology with the human, bovine, rat and mouse L/B/K-type ALP was found to be 88-90% at both the nucleotide and the amino acid levels. The cDNA was transferred into a eukaryotic expression vector and expressed following transfection into murine and mink lung fibroblast cell lines. High levels of enzymatically active ALP were detected, along with a 70-kDa **protein** reactive in immunoblot assay using a polyclonal antibody against the original crude FeALP preparation. FeALP was specifically released from intact cells by treatment with **phosphoinositol**-specific phospholipase-C. By Northern blot analysis, only one species of mRNA was detected using a 32P-labeled cDNA probe. These results indicate that the 2127-bp cDNA encodes a functional feline L/B/K-type ALP expressed on cell surfaces via phosphatidylinositol-glycan linkage. Despite electrophoretic comigration in two dimensions and following deglycosylation in a

third dimension, FeALP failed to function as an FeLV receptor since its expression failed to provide for attachment or entry of virus into cells.

L21 ANSWER 46 OF 59 MEDLINE DUPLICATE 17
ACCESSION NUMBER: 96116215 MEDLINE
DOCUMENT NUMBER: 96116215
TITLE: Structure and expression of an ovine endometrial oxytocin receptor cDNA.
AUTHOR: Riley P R; Flint A P; Abayasekara D R; Stewart H J
CORPORATE SOURCE: Institute of Zoology, Regents Park, London, UK.
SOURCE: JOURNAL OF MOLECULAR ENDOCRINOLOGY, (1995 Oct) 15 (2) 195-202.
Journal code: AEG. ISSN: 0952-5041.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-X87986
ENTRY MONTH: 199612
AB A sheep endometrial oxytocin receptor (OTR) cDNA (1.5 kb) was isolated from a lambda-ZAP library using a reverse transcription-**PCR** product probe generated from oestrous endometrial mRNA. The sheep OTR cDNA shared an overall similarity of 82% with human OTR cDNA, 85% with pig OTR cDNA and 76% with rat OTR cDNA. The encoded receptor was a 391 amino acid **polypeptide** 94% similar to human OTR, 94% similar to pig OTR and 93% similar to rat OTR. The sheep OTR contained two additional amino acids compared with human OTR which were located in the highly GC-rich third intracytoplasmic loop. This region is thought to be associated with G **protein** coupling and signal transduction. Expression of the cDNA in Cos-7 cells and measurement of oxytocin-induced **phosphoinositide** turnover confirmed that it coded for a functional product. The affinity of the expressed receptor was comparable with that observed for the in vivo receptor.

L21 ANSWER 47 OF 59 SCISEARCH COPYRIGHT 1999 ISI (R)
ACCESSION NUMBER: 95:130069 SCISEARCH
THE GENUINE ARTICLE: QF765
TITLE: MOLECULAR-CLONING OF THE PLC1(+) **GENE** OF SCHIZOSACCHAROMYCES-POMBE, WHICH ENCODES A PUTATIVE PHOSPHOINOSITIDE-SPECIFIC PHOSPHOLIPASE-C
AUTHOR: ANDOH T; YOKOO T; MATSUI Y; TOHE A (Reprint)
CORPORATE SOURCE: UNIV TOKYO, GRAD SCH SCI, DEPT PLANT SCI, BUNKYO KU, TOKYO 113, JAPAN (Reprint); UNIV TOKYO, GRAD SCH SCI, DEPT PLANT SCI, BUNKYO KU, TOKYO 113, JAPAN
COUNTRY OF AUTHOR: JAPAN
SOURCE: YEAST, (FEB 1995) Vol. 11, No. 2, pp. 179-185.
ISSN: 0749-503X.

DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE; AGRI
LANGUAGE: ENGLISH
REFERENCE COUNT: 31
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB Exploiting the **polymerase** chain **reaction**, we have isolated a **gene** that encodes a putative **phosphoinositide**-specific phospholipase C (PLC) of the fission yeast Schizosaccharomyces pombe. Inspection of the nucleotide sequence of the **gene** revealed an open reading frame that can encode a **polypeptide** of 899 amino acid residues with a calculated molecular mass of 102 kDa. This putative **polypeptide** contains both the X and Y regions that are conserved among three classes of mammalian PLC, and also contains a presumptive Ca2+-binding site (an E-F hand motif). The structure of the putative **protein** is most similar to that of the delta class of PLC isozymes. To investigate the role of this **gene**, designated plc1(+), **gene** disruption was carried out by interrupting the coding region with the ura4(+) marker. Growth of plc1 cells was temperature-sensitive in rich medium, and cells could not grow in synthetic medium. Expression of the PLC1 **gene** of Saccharomyces cerevisiae suppressed the growth defect phenotype of plc1(-) cells, a strong suggestion that the plc1(+) **gene** encodes PLC. The PLC1 sequence appears in the public data libraries, DDBJ GenBank, EMBL under the following Accession Number: D38309.

L21 ANSWER 48 OF 59 LIFESCI COPYRIGHT 1999 CSA
ACCESSION NUMBER: 94:77802 LIFESCI
TITLE: Cloning and expression of a novel, highly truncated phosphoinositide-specific phospholipase C cDNA from embryos of the brine shrimp, Artemia
AUTHOR: Su, Xilin; Chen, Fengling; Hokin, L.E.*
CORPORATE SOURCE: Dep. Pharmacol., Univ. Wisconsin Med. Sch., 1300 University Ave., Madison, WI 53706, USA
SOURCE: J. BIOL. CHEM., (1994) vol. 269, no. 17, pp. 12925-12931.
ISSN: 0021-9258.
DOCUMENT TYPE: Journal
FILE SEGMENT: Q4; N; G
LANGUAGE: English
SUMMARY LANGUAGE: English

AB A novel, highly truncated form of a cDNA encoding Artemia **phosphoinositide**-specific phospholipase C (PLC), designated PLC- beta sub(x), was isolated from a brine shrimp cDNA library. The full-length cDNA is of the beta -type, it is 2855 base pairs long, and it contains an open reading frame encoding 489 amino acids. The deduced amino acid sequence of PLC- beta sub(x) cDNA shows novel features. It lacks several hundred amino acids at the 5' end, as compared to PLC- beta s in the higher species. It contains

conserved domains X and Y, but domain X is highly truncated at the 5' end (only 14-25 conserved amino acids as compared to about 150 amino acids in the higher eukaryotic organisms). Northern blot hybridization showed that the PLC- beta sub(x) cDNA corresponds to a 4.4-kilobase mRNA. Northern blot hybridization with a cDNA probe from the 5' end and **PCR** performed upstream from domain Y showed that PLC- beta sub(x) is not a cloning artifact due to fusion of an unrelated clone into the coding region of the PLC- beta homologue.

L21 ANSWER 49 OF 59 MEDLINE
ACCESSION NUMBER: 94209312 MEDLINE
DOCUMENT NUMBER: 94209312
TITLE: Substitution of glutamine for arginine 1131. A newly identified mutation in the catalytic loop of the tyrosine kinase domain of the human insulin receptor.
AUTHOR: Kishimoto M; Hashiramoto M; Yonezawa K; Shii K; Kazumi T; Kasuga M
CORPORATE SOURCE: Second Department of Internal Medicine, Kobe University School of Medicine, Japan.
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (1994 Apr 15) 269 (15) 11349-55.
Journal code: HIV. ISSN: 0021-9258.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199407

AB We studied a patient with severe insulin resistance and a remarkable decrease in the in vivo autophosphorylation of the insulin receptor. Using a **polymerase** chain **reaction**-single strand conformation polymorphism method and direct sequencing, we identified a heterozygous mutation substituting Gln for Arg1131 in the putative "catalytic loop" of the tyrosine kinase domain of the insulin receptor **gene**. The Gln1131 mutant receptor was expressed by transfection in Chinese hamster ovary cells and compared with cells expressing the wild-type insulin receptor. Both mutant and wild-type receptors were expressed on the cell surface and displayed similar insulin-binding affinity. The Gln1131 mutation impaired the activity of the receptor tyrosine kinase and inhibited the ability of insulin to phosphorylate the endogenous substrate insulin receptor substrate-I. In addition, the Gln1131 mutant receptor exhibited diminished tyrosine-phosphorylated **phosphatidylinositol 3-kinase** and myelin basic **protein** kinase activities compared with the wild-type cells. It also demonstrated a defective mediation of the insulin signal stimulating 2-deoxy-D-glucose transport and thymidine incorporation, resistance to endocytosis, and insulin-induced down-regulation. Unlike a previously described mutation in the

putative catalytic loop of the receptor that substituted Glu for Ala1135, the Gln1131 mutation retained proteolytic cleavage of the proreceptor into separate subunits. Our results demonstrate that a naturally occurring mutation (R1131Q) in the putative catalytic loop of the insulin receptor results in severe impairment of the tyrosine kinase function in our patient. In addition, our results indicate that Arg1131 is important for receptor-mediated insulin action in vivo and suggest that the amino acids constituting the catalytic loop of **protein** kinases may possess different modes in order to retain kinase function.

L21 ANSWER 50 OF 59 SCISEARCH COPYRIGHT 1999 ISI (R)
ACCESSION NUMBER: 94:762748 SCISEARCH
THE GENUINE ARTICLE: PT844
TITLE: DELETION OF 3 BASEPAIRS RESULTING IN THE LOSS OF LYSINE-121 IN THE INSULIN-RECEPTOR ALPHA-SUBUNIT IN A PATIENT WITH LEPRECHAUNISM - BINDING, PHOSPHORYLATION, AND BIOLOGICAL-ACTIVITY
AUTHOR: JOSPE N (Reprint); ZHU J; LIU R C; LIVINGSTON J N; FURLANETTO R W
CORPORATE SOURCE: UNIV ROCHESTER, SCH MED & DENT, STRONG CHILDRENS MED CTR, DEPT PEDIAT, DIV PEDIAT ENDOCRINOL, ROCHESTER, NY, 14642 (Reprint); UNIV ROCHESTER, SCH MED & DENT, DEPT BIOCHEM, ROCHESTER, NY, 14642; MILES INC, INST METAB DISORDERS, W HAVEN, CT, 06516
COUNTRY OF AUTHOR: USA
SOURCE: JOURNAL OF CLINICAL ENDOCRINOLOGY AND METABOLISM, (NOV 1994) Vol. 79, No. 5, pp. 1294-1302. ISSN: 0021-972X.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE; CLIN
LANGUAGE: ENGLISH
REFERENCE COUNT: 35
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB We have identified a novel mutation of the human insulin receptor **gene** in a previously unreported patient with leprechaunism, leprechaun Rochester. This mutation consists of deletion of three nucleotides (GAA) in exon 2 and results in loss of the lysine-121 in the putative ligand-binding domain of the Lu-subunit. To analyze this mutation, we prepared a corresponding mutant insulin receptor by site-directed mutagenesis and expressed the receptor in Chinese hamster ovary cells. Although the mutant receptor displayed normal insulin binding, abnormalities were found in autophosphorylation and in phosphorylation of endogenous and exogenous **protein** substrates. These abnormalities consisted of increased basal kinase activity, but blunted insulin-stimulated responsiveness. Importantly, cells that expressed the mutant receptor showed markedly decreased insulin- and serum-stimulated **DNA** synthesis compared to untransfected control cells and cells

transfected with the wild-type insulin receptor. These findings suggest that deletion of lysine-121 in conjunction with a presumed but thus far unidentified, second mutant allele contributed significantly to the lethal insulin-resistant state in this patient with leprechaunism.

L21 ANSWER 51 OF 59 EMBASE COPYRIGHT 1999 ELSEVIER SCI. B.V.DUPLICATE 18

ACCESSION NUMBER: 95021694 EMBASE
DOCUMENT NUMBER: 1995021694
TITLE: Molecular cloning, cDNA sequence, and chromosomal localization of the human phosphatidylinositol 3-kinase p110.alpha. (PIK3CA) **gene**.
AUTHOR: Volinia S.; Hiles I.; Ormondroyd E.; Nizetic D.; Antonacci R.; Rocchi M.; Waterfield M.D.
CORPORATE SOURCE: Ludwig Institute for Cancer Research, 91 Riding House Street,London W1P 8BT, United Kingdom
SOURCE: Genomics, (1994) 24/3 (472-477).
ISSN: 0888-7543 CODEN: GNMCEP
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 022 Human Genetics
029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Phosphatidylinositol (PI) 3-kinase is a heterodimeric enzyme comprising a 110-kDa catalytic subunit and an 85-kDa regulatory subunit that binds to tyrosine phosphopeptide sites linked directly or indirectly to receptors serving diverse signal functions. Knowledge of the structure and function of PI 3-kinase was greatly advanced by the purification, cDNA cloning, and subsequent expression of the bovine enzyme. Here the cloning of the cDNA for the human p110.alpha. subunit of **PI3-kinase** (PLK3CA), encoding a **protein** 99% identical to the bovine p110, and of its **gene** in YAC is described. The chromosomal localization of the **gene** for PIK3CA is shown to be at 3q21-qter as determined using somatic cell hybrids. In situ hybridization performed using Alu-**PCR** from the YAC **DNA** located the **gene** in 3q26.3.

L21 ANSWER 52 OF 59 MEDLINE DUPLICATE 19

ACCESSION NUMBER: 94131004 MEDLINE
DOCUMENT NUMBER: 94131004
TITLE: Expression of three alternative acetylcholinesterase messenger **RNAs** in human tumor cell lines of different tissue origins.
AUTHOR: Karpel R; Ben Aziz-Aloya R; Sternfeld M; Ehrlich G; Ginzberg D; Tarroni P; Clementi F; Zakut H; Soreq H
CORPORATE SOURCE: Department of Biological Chemistry, Hebrew

University, Jerusalem, Israel.
SOURCE: EXPERIMENTAL CELL RESEARCH, (1994 Feb) 210 (2) 268-77.
Journal code: EPB. ISSN: 0014-4827.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199405

AB To study the molecular mechanisms underlying the intensive expression of acetylcholinesterase (AChE) in different tumor types, we characterized levels and composition of its messenger **RNA** (mRNA) sequences in heterologous tumor cell lines, primary tumor biopsies, and normal fetal and adult tissues and determined their exon-intron origin within the corresponding ACHE **gene**. Reverse transcription followed by **polymerase** chain **reaction** (RT-**PCR**) revealed three alternatively spliced ACHE mRNAs in NT2/D1 teratocarcinoma, NCI-N-592 small cell lung carcinoma, TE671 medulloblastoma, K-562 erythroleukemia, and 293 transformed embryonal kidney cells. The three ACHE mRNAs include the principal species expressed in brain and muscle and two additional transcripts containing insertions of 751 or 829 residues downstream from the exon 4 domain. The inserted region, which represents an intron in brain and muscle, is expressed in the tumor cell lines either as a "readthrough" form or with 78 residues deleted from its 5' end. A major band of 2.5 kb was labeled with ACHE cDNA in poly(A)+ **RNA** blots from medulloblastoma cells or brain tissue, whereas a **PCR**-amplified probe from the inserted domain labeled a 3.4-kb band but not the 2.5-kb band in poly(A)+ **RNA** from small cell lung carcinoma. The ACHE mRNAs including the alternative insertions were found only in cell lines with levels of the principal ACHE mRNA species equal to or higher than those in brain (1-10 molecules/cell), determined by following the kinetics of mRNA **PCR** amplification. Genomic **DNA** sequencing revealed that the inserted domains in the ACHE mRNAs expressed in the tumor cell lines encode C-terminal **peptides** of 40 and 14 residues. These include a free cysteine, terminate with the consensus HG element, and continue by a 29-residue-long C-terminal hydrophobic cleavable **peptide**, properties characteristic of precursors to **phosphoinositide** (PI)-linked **proteins**. In extension of the reported expression of PI-linked AChE in hemopoietic cells including K-562, our findings demonstrate the existence of ACHE mRNAs with the potential to encode one hydrophilic and two PI-linked forms of AChE in tumor cells from both hemopoietic and nonhemopoietic origins.

L21 ANSWER 53 OF 59 JICST-EPlus COPYRIGHT 1999 JST
ACCESSION NUMBER: 940533357 JICST-EPlus
TITLE: Studies on Biological Implication of

Gonadotropin-releasing Hormone(Gn-RH) and its Receptor in Human Ovarian Carcinoma.

AUTHOR: ONO TSUKASA; IMAI ATSUSHI; TAMAYA TERUHIKO
CORPORATE SOURCE: Gifu Univ., Sch. of Med.

SOURCE: Gifu Daigaku Igakubu Kiyo (Acta Scholae Medicinalis Universitatis in Gifu), (1994) vol. 42, no. 2, pp. 174-185. Journal Code: F0639A (Fig. 10, Ref. 55)
CODEN: GDIKAN; ISSN: 0072-4521
PUB. COUNTRY: Japan
DOCUMENT TYPE: Journal; Article
LANGUAGE: Japanese
STATUS: New

AB This study was designed to investigate the biological implication of gonadotropin-releasing hormone(Gn-RH) and its receptor in human ovarian carcinoma, in order to know the possible clinical use of Gn-RH analogs in the treatment of ovarian carcinoma. Gn-RH was determined by a radioimmunoassay and bioassay. Gn-RH messenger **ribonucleic** acid(mRNA) was determined by reverse transcription(RT)-**polymerase** chain **reaction**(**PCR**) using oligonucleotide primers synthesized according to the published Gn-RH **gene** sequence. 1) Gn-RH was shown to be present in extracts of ovarian mucinous cystadenocarcinoma samples (0.8.+-.0.12pg/mg **protein**) and ovarian adenocarcinoma cell line SK-OV-3 (0.92.+-.0.17pg/mg **protein**), but not in normal ovary and placenta. 2) Two of two extract samples from individual cases of mucinous cystadenocarcinoma evoked dose-dependent **phosphoinositide** breakdown in rat granulosa cells, similar to that caused by authentic Gn-RH. 3) Gn-RH mRNA was detected in two of two mucinous cystadenocarcinoma specimens, one of one serous cystadenocarcinoma, and SK-OV-3 cells, but not in mucinous cystadenoma, the normal ovary and placenta. Gn-RH receptor was determined by the presences of .cents.3H!Gn-RH binding sites and its mRNA by RT-**PCR** method. The transmembrane signaling events interacting with Gn-RH receptor were also established in **phosphoinositide** turnover. 1) Specific Gn-RH binding sites were shown to be present in the plasma membrane isolated from two ovarian mucinous cystadenocarcinoma samples (Kd=8.0nM, Bmax=0.1 to 0.2pmol/mg **protein**). 2) Gn-RH receptor mRNA was detected in two of two mucinous cystadenocarcinoma specimens, one of one serous cystadenocarcinoma, and SK-OV-3 cells, but not in female white blood cells. 3) In mucinous cystadenocarcinoma, incubation of plasma membranes isolated from .cents.3H! inositol-labelled specimens with Gn-RH resulted in the rapid production of inositol phosphates. (abridged author abst.)

L21 ANSWER 54 OF 59 MEDLINE DUPLICATE 20
ACCESSION NUMBER: 94131738 MEDLINE
DOCUMENT NUMBER: 94131738
TITLE: Endothelin-mediated cell signaling and proliferation

in cultured rabbit corneal epithelial cells.

AUTHOR: Takagi H; Reinach P S; Tachado S D; Yoshimura N
CORPORATE SOURCE: Department of Ophthalmology, Kyoto University Faculty of Medicine, Japan.

CONTRACT NUMBER: EY04795 (NEI)
SOURCE: INVESTIGATIVE OPHTHALMOLOGY AND VISUAL SCIENCE, (1994 Jan) 35 (1) 134-42.
Journal code: GWI. ISSN: 0146-0404.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199405

AB PURPOSE. To determine if there is endothelin-mediated regulation of cell signaling and proliferation in rabbit corneal epithelium. METHODS. Endothelin-1 (ET-1) **gene** and **protein** expression by the rabbit corneal epithelial (RCE) cells were analyzed by **polymerase** chain **reaction**, sequence analysis, and enzyme immunoassay. **DNA** synthesis was characterized by [3H]-thymidine uptake. Endothelin receptor linkage to cell signaling pathways was determined based on measurements of the dose dependent effects of ET-1, ET-2, and ET-3 on intracellular Ca2+ concentration ([Ca2+]i) transients in fura-2-loaded cells, and of ET-1 on **phosphoinositide** turnover and cAMP accumulation in the isolated rabbit corneal epithelium. RESULTS. The authors detected the mRNA for prepro ET-1 in RCE cells, and ET-like immunoreactivity was identified in conditioned culture medium. ET-1 (1 nM) maximally stimulated [3H]-thymidine uptake by twofold (EC50 = 0.3 nM). Endothelins elicited transient increases in [Ca2+]i with a rank order of potency of ET-1 > or = ET-2 >> ET-3. These increases consisted of both intracellular Ca2+ mobilization and influx of Ca2+ from the bathing solution. Intracellular mobilization was linked to increases in IP3 turnover because 1 microM ET-1 increased IP3 content by 48% from its control value (EC50 = 23 nM), whereas Ca2+ influx occurred through a non-L-type Ca2+ channel because preexposure to 1 microM nicardipine did not affect either the height or the duration of a [Ca2+]i transient. One micromolar of ET-1 was required to elicit a significant increase in cAMP accumulation of 69% from its control value. This increase was dependent on the presence of Ca2+ in the bathing solution and was comparable to and nonadditive with that of the Ca2+ ionophore, A23187 (1 microM). CONCLUSION. These data suggest that endothelin production by primary cultures of RCE cells can mediate an increase in cell proliferation through an ETA receptor subtype. This receptor subtype appears to be involved based on the rank order of potency of ETs to elicit [Ca2+]i transients, increases in **phosphoinositide** turnover, and cAMP accumulation.

ACCESSION NUMBER: 93:148390 SCISEARCH
THE GENUINE ARTICLE: KP979
TITLE: THE PUTATIVE PHOSPHOINOSITIDE-SPECIFIC PHOSPHOLIPASE-C **GENE**, PLC1, OF THE YEAST SACCHAROMYCES-CEREVISIAE IS IMPORTANT FOR CELL-GROWTH
AUTHOR: YOKOO T; MATSUI Y; YAGISAWA H; NOJIMA H; UNO I; TOHE A (Reprint)
CORPORATE SOURCE: UNIV TOKYO, FAC SCI, DEPT BIOL, BUNKYO KU, TOKYO 113, JAPAN; HIMEJI INST TECHNOL, FAC SCI, DEPT LIFE SCI, KAMIGORI, HYOGO 67812, JAPAN; RES INST MICROBIAL DIS, DEPT MOLEC GENET, SUITA, OSAKA 565, JAPAN; NIPPON STEEL CORP LTD, RES & DEV LABS 1, KAWASAKI 211, JAPAN
COUNTRY OF AUTHOR: JAPAN
SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (01 MAR 1993) Vol. 90, No. 5, pp. 1804-1808.
ISSN: 0027-8424.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE
LANGUAGE: ENGLISH
REFERENCE COUNT: 35
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB Using the **polymerase** chain **reaction** technique, we have isolated a **gene** that encodes a putative **phosphoinositide**-specific phospholipase C (PLC) in the yeast Saccharomyces cerevisiae. The nucleotide sequence indicates that the **gene** encodes a **polypeptide** of 869 amino acid residues with a calculated molecular mass of 101 kDa. This **polypeptide** has both the X and Y regions conserved among mammalian PLC-beta, -gamma, and -delta, and the structure is most similar to that of mammalian PLC-delta. This putative yeast PLC **gene** has been designated PLC1. Disruption of PLC1 results in slow growth or lethality for cells, depending on their genetic background and the medium, indicating that PLC1 is important for cell growth. Expression of rat PLC-delta1 cDNA suppressed the growth defect of plc1 disruptants, strongly suggesting that PLC1 encodes PLC.

L21 ANSWER 56 OF 59 MEDLINE
ACCESSION NUMBER: 94013473 MEDLINE
DOCUMENT NUMBER: 94013473
TITLE: Identification of mutations in the coding sequence of the proto-oncogene c-kit in a human mast cell leukemia cell line causing ligand-independent activation of c-kit product.
AUTHOR: Furitsu T; Tsujimura T; Tono T; Ikeda H; Kitayama H; Koshimizu U; Sugahara H; Butterfield J H; Ashman L K;

Kanayama Y; et al
CORPORATE SOURCE: Second Department of Internal Medicine, Osaka University Medical School, Suita, Japan.
SOURCE: JOURNAL OF CLINICAL INVESTIGATION, (1993 Oct) 92 (4) 1736-44.
Journal code: HS7. ISSN: 0021-9738.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals; Cancer Journals
ENTRY MONTH: 199401
AB The c-kit proto-oncogene encodes a receptor tyrosine kinase. Binding of c-kit ligand, stem cell factor (SCF) to c-kit receptor (c-kitR) is known to activate c-kitR tyrosine kinase, thereby leading to autophosphorylation of c-kitR on tyrosine and to association of c-kitR with substrates such as **phosphatidylinositol 3-kinase** (PI3K). In a human mast cell leukemia cell line HMC-1, c-kitR was found to be constitutively phosphorylated on tyrosine, activated, and associated with PI3K without the addition of SCF. The expression of SCF mRNA transcript in HMC-1 cells was not detectable by means of **PCR** after reverse transcription (RT-**PCR**) analysis, suggesting that the constitutive activation of c-kitR was ligand independent. Sequencing of whole coding region of c-kit cDNA revealed that c-kit **genes** of HMC-1 cells were composed of a normal, wild-type allele and a mutant allele with two point mutations resulting in intracellular amino acid substitutions of Gly-560 for Val and Val-816 for Asp. Amino acid sequences in the regions of the two mutations are completely conserved in all of mouse, rat, and human c-kit. In order to determine the causal role of these mutations in the constitutive activation, murine c-kit mutants encoding Gly-559 and/or Val-814, corresponding to human Gly-560 and/or Val-816, were constructed by site-directed mutagenesis and expressed in a human embryonic kidney cell line, 293T cells. In the transfected cells, both c-kitR (Gly-559, Val-814) and c-kitR (Val-814) were abundantly phosphorylated on tyrosine and activated in immune complex kinase reaction in the absence of SCF, whereas tyrosine phosphorylation and activation of c-kitR (Gly-559) or wild-type c-kitR was modest or little, respectively. These results suggest that conversion of Asp-816 to Val in human c-kitR may be an activating mutation and responsible for the constitutive activation of c-kitR in HMC-1 cells.

L21 ANSWER 57 OF 59 SCISEARCH COPYRIGHT 1999 ISI (R)
ACCESSION NUMBER: 93:582386 SCISEARCH
THE GENUINE ARTICLE: LY025
TITLE: PRESENCE OF GONADOTROPIN-RELEASING-HORMONE AND ITS MESSENGER-**RIBONUCLEIC**-ACID IN HUMAN

OVARIAN EPITHELIAL CARCINOMA

AUTHOR: OHNO T; IMAI A (Reprint); FURUI T; TAKAHASHI K; TAMAYA T
CORPORATE SOURCE: GIFU UNIV, SCH MED, DEPT OBSTET & GYNECOL, GIFU 500, JAPAN
COUNTRY OF AUTHOR: JAPAN
SOURCE: AMERICAN JOURNAL OF OBSTETRICS AND GYNECOLOGY, (SEP 1993) Vol. 169, No. 3, pp. 605-610.
ISSN: 0002-9378.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE; CLIN
LANGUAGE: ENGLISH
REFERENCE COUNT: 36

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB OBJECTIVE: The purpose of this study was to investigate the expression of gonadotropin-releasing hormone messenger **ribonucleic** acid and the presence of gonadotropin-releasing hormone in human ovarian carcinoma known to have gonadotropin-releasing hormone binding sites and to be affected by gonadotropin-releasing hormone analog.

STUDY DESIGN: Human ovarian carcinomas surgically removed and human ovarian carcinoma cell lines were examined. Gonadotropin-releasing hormone was determined by a radioimmunoassay and a bioassy. Gonadotropin-releasing hormone messenger **ribonucleic** acid was determined by reverse transcription **polymerase** chain **reaction** using oligonucleotide primers synthesized according to the published human gonadotropin-releasing hormone sequence.

RESULTS: Gonadotropin-releasing hormone was shown to be present in extracts of ovarian mucinous cystadenocarcinoma sample (0.8 +/- 0.12 pg/mg of **protein**) and ovarian adenocarcinoma cell line SK-OV3 (0.92 +/- 0.17 pg/mg of **protein**) but not in the normal ovary and placenta. Two of two extract samples from individual cases evoked dose-dependent **phosphoinositide** breakdown in rat granulosa cells similar to that caused by authentic gonadotropin-releasing hormone, Gonadotropin-releasing hormone messenger **ribonucleic** acid was detected in two of two mucinous cystadenocarcinoma specimens, one of one serous cystadenocarcinoma, and SK-OV3 cells but not in the dysgerminoma, mucinous cystadenoma, and normal ovary and placenta.

CONCLUSION: The demonstration of gonadotropin-releasing hormone and its messenger **ribonucleic** acid raises the possibility that gonadotropin-releasing hormone may play an autocrine regulatory role in the growth of ovarian carcinoma.

L21 ANSWER 58 OF 59 MEDLINE DUPLICATE 21
ACCESSION NUMBER: 92406741 MEDLINE
DOCUMENT NUMBER: 92406741
TITLE: Molecular cloning and expression of a

phosphoinositide-specific phospholipase C of Dictyostelium discoideum.
AUTHOR: Drayer A L; van Haastert P J
CORPORATE SOURCE: Department of Biochemistry, University of Groningen, The Netherlands..
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (1992 Sep 15) 267 (26) 18387-92.
Journal code: HIV. ISSN: 0021-9258.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
OTHER SOURCE: GENBANK-M95783; GENBANK-D10868; GENBANK-D10869; GENBANK-D10870; GENBANK-D10871; GENBANK-D10872; GENBANK-D10345; GENBANK-D10346; GENBANK-D10347; GENBANK-D10348
ENTRY MONTH: 199212

AB A number of **phosphoinositide**-specific phospholipases C (PLC) of different species have recently been cloned. The predicted amino acid sequences of these isoforms contain two highly conserved domains. Here we report the identification of a PLC **gene** of Dictyostelium by using the **polymerase** chain **reaction**. Primers were designed coding for highly conserved amino acid regions located within one of the conserved domains of PLCs. Cloning and sequencing of the **polymerase** chain **reaction** product revealed one unique PLC-like sequence. This sequence was used to screen a library and isolate several overlapping cDNA clones. The complete cDNA was expressed in Dictyostelium cells resulting in increased basal levels of inositol 1,4,5-trisphosphate and enhanced PLC activity. The identified Dictyostelium PLC, DdPLC, encodes a **protein** with a calculated molecular mass of 91 kDa. The deduced amino acid sequence contains the two conserved domains found in other PLC isoforms, separated by a short variable region. The C-terminal part of the **protein** shows strong homology with the mammalian PLC-delta isoform. DdPLC is expressed at all stages of development, with an increase in transcription during starvation and in the culminating fruiting body.

L21 ANSWER 59 OF 59 SCISEARCH COPYRIGHT 1999 ISI (R)
ACCESSION NUMBER: 91:648112 SCISEARCH
THE GENUINE ARTICLE: GR061
TITLE: PHOSPHOLIPASE C-DELTA **GENE** OF THE SPONTANEOUSLY HYPERTENSIVE RAT HARBORS POINT MUTATIONS CAUSING AMINO-ACID SUBSTITUTIONS IN A CATALYTIC DOMAIN
AUTHOR: YAGISAWA H; TANASE H; NOJIMA H (Reprint)
CORPORATE SOURCE: OSAKA UNIV, MICROBIAL DIS RES INST, DEPT MOLEC GENET, SUITA, OSAKA 565, JAPAN; SANKYO CO LTD, LAB

ANIM SCI & TOXICOL LABS, FUKUROI, SHIZUOKA, JAPAN; JICHI MED SCH, DEPT MED BIOL & PARASITOL, MINAMI KAWACHI, TOCHIGI 32904, JAPAN
COUNTRY OF AUTHOR: JAPAN
SOURCE: JOURNAL OF HYPERTENSION, (1991) Vol. 9, No. 11, pp. 997-1004.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE
LANGUAGE: ENGLISH
REFERENCE COUNT: 32

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB This study was undertaken in order to investigate the newly discovered spontaneously hypertensive rat (SHR)-specific restriction fragment length polymorphism (RFLP) at the genomic locus of (poly) **phosphoinositide**-specific phospholipase C (PLC)-delta at a **DNA** sequence level. Our aim was to clone the PLC-delta complimentary **DNA** (cDNA) from SHR and analyse the genomic **DNA** obtained from two hypertensive rat strains such as SHR and its stroke-prone substrain (SHR-SP) and three normotensive rat strains such as Sprague-Dawley, Donryu and Wistar-Kyoto (WKY) by preparing an aortic cDNA library of SHR, hybridization cloning of PLC-delta cDNA and an analysis of the genomic **DNA** by **polymerase** chain **reaction**. By digesting with restriction enzyme XhoI, we discovered an RFLP band displaying only in SHR and SHR-SP, not in Sprague-Dawley, Donryu and WKY rats. **DNA** sequencing of PLC-delta cDNA cloned from an aortic cDNA library of SHR revealed a total of three SHR-specific point mutations, two of which resulted in amino acid substitutions. The first point mutation (A to T) was detected at the XhoI site, changing a threonine(ACG) to a serine(TCG), and the second point mutation (A to G) was discovered in the vicinity of the first one, changing an isoleucine(ATA) to a methionine(ATG). This is the first demonstration of the mutations in the SHR genome changing amino acid sequences. These amino acid substitutions, situated in the putative catalytic X domain of PLC-delta, may be the major cause of the augmented PLC activity observed in the SHR, possibly leading to hypertension-related phenonemona such as abnormal calcium homeostasis and increased intracellular calcium ion concentrations.

=> d his l22- ful; d 1-5 ibib abs

(FILE 'CAPLUS, MEDLINE, BIOSIS, EMBASE, LIFESCI, BIOTECHDS, WPIDS, CONFSCI, SCISEARCH, JICST-EPLUS, PROMT' ENTERED AT 14:37:41 ON 16 SEP 1999)

L22 139 SEA ABB=ON PLU=ON HILES I?/AU
L23 967 SEA ABB=ON PLU=ON FRY M?/AU
L24 328 SEA ABB=ON PLU=ON DHAND R?/AU
L25 1208 SEA ABB=ON PLU=ON WATERFIELD M?/AU
L26 3303 SEA ABB=ON PLU=ON PARKER P?/AU

_ Authors

Searcher : Shears 308-4994

L27 918 SEA ABB=ON PLU=ON OTSU M?/AU
L28 320 SEA ABB=ON PLU=ON PANAYOTOU G?/AU
L29 231 SEA ABB=ON PLU=ON VOLINIA S?/AU
L30 225 SEA ABB=ON PLU=ON GOUT I?/AU
L31 4 SEA ABB=ON PLU=ON L22 AND L23 AND L24 AND L25 AND L26 AND L27 AND L28 AND L29 AND L30
L32 86 SEA ABB=ON PLU=ON L22 AND (L23 OR L24 OR L25 OR L26 OR L27 OR L28 OR L29 OR L30)
L33 130 SEA ABB=ON PLU=ON L23 AND (L24 OR L25 OR L26 OR L27 OR L28 OR L29 OR L30)
L34 114 SEA ABB=ON PLU=ON L24 AND (L25 OR L26 OR L27 OR L28 OR L29 OR L30)
L35 432 SEA ABB=ON PLU=ON L25 AND (L26 OR L27 OR L28 OR L29 OR L30)
L36 19 SEA ABB=ON PLU=ON L26 AND (L27 OR L28 OR L29 OR L30)
L37 27 SEA ABB=ON PLU=ON L27 AND (L28 OR L29 OR L30)
L38 72 SEA ABB=ON PLU=ON L28 AND (L29 OR L30)
L39 28 SEA ABB=ON PLU=ON L29 AND L30
L40 279 SEA ABB=ON PLU=ON (L32 OR L33 OR L34 OR L35 OR L36 OR L37 OR L38 OR L39) AND L2
L41 279 SEA ABB=ON PLU=ON L31 OR L40
L42 77 DUP REM L41 (202 DUPLICATES REMOVED)
L43 14 SEA ABB=ON PLU=ON L40 AND (POLYMERASE(1W) REACT? OR PCR)
L44 18 SEA ABB=ON PLU=ON L31 OR L43
L45 5 DUP REM L44 (13 DUPLICATES REMOVED)

L45 ANSWER 1 OF 5 BIOSIS COPYRIGHT 1999 BIOSIS
ACCESSION NUMBER: 1999:28052 BIOSIS
DOCUMENT NUMBER: PREV199900028052
TITLE: Human **phosphoinositide** 3-kinase C2beta, the role of calcium and the C2 domain in enzyme activity.
AUTHOR(S): Arcaro, Alexandre; **Volinia, Stefano**; Zvelebil, Marketa J.; Stein, Rob; Watton, Sandra J.; Layton, Meredith J.; **Gout, Ivan**; Ahmadi, Khatereh; Downward, Julian; **Waterfield, Michael D. (1)**
CORPORATE SOURCE: (1) Ludwig Inst. Cancer Res., University Coll. London, 91 Riding House St., London W1P 8BT UK
SOURCE: Journal of Biological Chemistry, (Dec. 4, 1998) Vol. 273, No. 49, pp. 33082-33090.
ISSN: 0021-9258.
DOCUMENT TYPE: Article
LANGUAGE: English
AB The cDNA for a human Class II **phosphoinositide** 3-kinase (PI 3-kinase C2beta) with a C2 domain was cloned from a U937 monocyte cDNA library and the enzyme expressed in mammalian and insect cells. Like other Class II PI 3-kinases in vitro, PI 3-kinase C2beta utilizes phosphatidylinositol (PI) and PI 4-monophosphate but

not PI 4,5-biphosphate as substrates in the presence of Mg2+. Remarkably, and unlike other PI 3-kinases, the enzyme can use either Mg-ATP or Ca-ATP to generate PI 3-monophosphate. PI 3-kinase C2beta, like the Class I PI 3-kinases, but unlike PI 3-kinase C2alpha, is sensitive to low nanomolar levels of the inhibitor wortmannin. The enzyme is not regulated by the small GTP-binding protein Ras. The C2 domain of the enzyme bound anionic phospholipids such as PI and phosphatidylserine in vitro, but did not co-operatively bind Ca2+ and phospholipids. Deletion of the C2 domain increased the lipid kinase activity suggesting that it functions as a negative regulator of the catalytic domain. Although presently it is not known whether PI 3-kinase C2beta is regulated by Ca2+ in vivo, our results suggest a novel role for Ca2+ ions in phosphate transfer reactions.

L45 ANSWER 2 OF 5 CAPLUS COPYRIGHT 1999 ACS DUPLICATE 1
ACCESSION NUMBER: 1995:303601 CAPLUS
DOCUMENT NUMBER: 122:206627
TITLE: Molecular cloning, cDNA sequence, and chromosomal localization of the human **phosphatidylinositol 3-kinase** p110.alpha. (PIK3CA) gene
AUTHOR(S): **Volinia, Stefano; Hiles, Ian** ; Ormondroyd, Elizabeth; Nizetic, Dean; Antonacci, Rachele; Rocchi, Mariano; **Waterfield, Michael D.**
CORPORATE SOURCE: Ludwig Inst. Cancer Res., London, W1P 8BT, UK
SOURCE: Genomics (1994), 24(3), 472-7
CODEN: GNMCEP; ISSN: 0888-7543
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Phosphatidylinositol (PI) 3-kinase is a heterodimeric enzyme comprising a 110-kDa catalytic subunit and an 85-kDa regulatory subunit that binds to tyrosine phosphopeptide sites linked directly or indirectly to receptors serving diverse signal functions. Knowledge of the structure and function of PI 3-kinase was greatly advanced by the purifn., cDNA cloning, and subsequent expression of the bovine enzyme. Here, the cloning of the cDNA for the human p110.alpha. subunit of **PI3-kinase** (PIK3CA), encoding a protein 99% identical to the bovine p110, and of its gene in YAC is described. The chromosomal localization of the gene for PIK3CA is shown to be at 3q21-qter as detd. using somatic cell hybrids. In situ hybridization performed using Alu-**PCR** from the YAC DNA located the gene in 3q26.3.

L45 ANSWER 3 OF 5 CAPLUS COPYRIGHT 1999 ACS DUPLICATE 2
ACCESSION NUMBER: 1994:100544 CAPLUS
DOCUMENT NUMBER: 120:100544
TITLE: Cloning and expression of a cDNA for a subunit of PI3 kinase

INVENTOR(S): **Hiles, Ian D.; Fry, Michael J. ; Dhand, Ritu; Waterfield, Michael D.; Parker, Peter J.; Otsu, Masayuki; Panayotou, George; Volinia, Stefano; Gout, Ivan**
PATENT ASSIGNEE(S): Ludwig Institute for Cancer Research, Barbados
SOURCE: PCT Int. Appl., 146 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9321328	A1	19931028	WO 1993-GB761	19930413
W: AU, CA, JP, US				
RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
AU 9339017	A1	19931118	AU 1993-39017	19930413
AU 664893	B2	19951207		
EP 590126	A1	19940406	EP 1993-908028	19930413
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LI, LU, MC, NL, PT, SE				
JP 06510207	T2	19941117	JP 1993-518112	19930413
JP 07203963	A2	19950808	JP 1994-4313	19940120
US 5824492	A	19981020	US 1994-162081	19940207
US 5846824	A	19981208	US 1997-780872	19970109
PRIORITY APPLN. INFO.:			GB 1992-8135	19920413
			WO 1993-GB761	19930413
			US 1994-162081	19940207

AB PI3 kinase activity is manufd. by expression of the cloned genes in insect cell culture. Affinity purifn. of the protein from bovine brain using Y751 phosphopeptide from PDGF-.beta. receptor as the affinity ligand identified the p85 and p110 (actual mol. wt. 124 kDa) proteins that bound to the column with very high affinity and appeared to form a complex. A cDNA bank from SGBAF-1 cells in .lambda.Uni-ZAP was screened with amino acid sequence-derived oligonucleotide probes from the p110 protein. and the cDNA expressed in Sf9 cells using a baculovirus vector. A cDNA clone was expressed in Sf9 cells using a baculovirus vector to give a detectable PI3 kinase activity. Tissue specificity of gene expression is detd. and the corresponding human cDNA was cloned.

L45 ANSWER 4 OF 5 CAPLUS COPYRIGHT 1999 ACS DUPLICATE 3
ACCESSION NUMBER: 1992:402084 CAPLUS
DOCUMENT NUMBER: 117:2084
TITLE: Chromosomal localization of human p85.alpha., a

subunit of **phosphatidylinositol 3-kinase**, and its homolog p85.beta.

AUTHOR(S): **Volinia, S.**; Patracchini, P.; **Otsu, M.**; **Hiles, I.**; **Gout, I.**; Calzolari, E.; Bernardi, F.; Rooke, L.; **Waterfield, M. D.**

CORPORATE SOURCE: Ludwig Inst. Cancer Res., London, W1P 8BT, UK

SOURCE: Oncogene (1992), 7(4), 789-93
CODEN: ONCNES; ISSN: 0950-9232

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The human phosphatidylinositol (PI) 3-kinase p85.alpha. subunit gene and its homolog p85.beta. were assigned to human chromosomes by anal. of their segregation in a panel of somatic cell hybrids using human-specific **PCR** primers. The p85.alpha. locus was only present in hybrids retaining the human chromosome 5q. The presence of the p85.beta. locus coincided with the presence of chromosome 19. The precise chromosomal sublocalization of these 2 genes was then detd. by in situ hybridization. The localization of the p58.alpha. gene was confirmed at 5q12-q13, as recently described (Cannizzaro, L. A., et al., 1991) and the p85.beta. locus positioned at 19q13.2-q13.4.

L45 ANSWER 5 OF 5 MEDLINE DUPLICATE 4

ACCESSION NUMBER: 92354059 MEDLINE

DOCUMENT NUMBER: 92354059

TITLE: **Phosphatidylinositol 3-kinase**: structure and expression of the 110 kd catalytic subunit.

AUTHOR: **Hiles I D**; **Otsu M**; **Volinia S**; **Fry M J**; **Gout I**; **Dhand R**; **Panayotou G**; Ruiz-Larrea F; Thompson A; Totty N F; et al

CORPORATE SOURCE: Ludwig Institute for Cancer Research, London, England.

SOURCE: CELL, (1992 Aug 7) 70 (3) 419-29.
Journal code: CQ4. ISSN: 0092-8674.

PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals; Cancer Journals

OTHER SOURCE: GENBANK-M93252; GENBANK-M94130; GENBANK-M94264; GENBANK-L05499; GENBANK-M87278; GENBANK-S72766; GENBANK-S72767; GENBANK-S72768; GENBANK-S72769; GENBANK-S72771

ENTRY MONTH: 199211

AB Purified bovine brain **phosphatidylinositol 3-kinase** (P13-kinase) is composed of 85 kd and 110 kd